HARVARD CASE HISTORIES IN EXPERIMENTAL SCIENCE
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CASE 7

Pasteur's and Tyndall's Study of Spontaneous Generation

EDITED BY

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Foreword

The Harvard Case Histories in Experimental Science have been designed primarily for students who are majoring in the humanities or the social sciences. Such students require an understanding of science that will help them to relate developments in the natural sciences to those in the other fields of human activity. To do so demands an understanding both of the methods of experimental science and of the growth of scientific research as an organized activity of society. Experience shows that a man who has been a successful investigator in any field of experimental science approaches a problem in pure or applied science, even in an area in which he is quite ignorant, with a special point of view. One may designate this point of view "understanding science"; it is independent of a knowledge of the scientific facts or techniques in the new area. Even a highly educated and intelligent citizen without research experience will almost always fail to grasp the essentials in a discussion that takes place among scientists concerned with a projected inquiry. This will be so not because of the layman's lack of scientific knowledge or his failure to comprehend the technical jargon of the scientist; it will be to a large degree because of his fundamental ignorance of what science can or cannot accomplish, and his subsequent bewilderment in the course of a discussion outlining a plan for a future investigation. He has no "feel" for what we may call "the tactics and strategy of science."

As a citizen, a businessman, a public servant, a lawyer, a teacher, or a writer, any present college student may a decade hence be called upon to evaluate the work of scientists and to consider the ways in which such work can be organized and financed. He may wish also to consider to what degree systematic investigations more or less analogous to those that have been successful in the physical sciences may be fruitful in other fields. Although the first cases are concerned almost exclusively with the physical sciences, an understanding of the variety of methods by which these sciences have advanced will provide some basis, it is hoped, from which the nonscientists can appraise proposals for research and its applications in the biological and social sciences.

1. UNDERSTANDING SCIENCE

In order to appreciate the methods of the experimental sciences it is necessary to master a certain amount of technical subject matter within the fields of physics and chemistry although no one can prescribe the particular topics to be studied. It is quite clear that one need not attempt the task of surveying all physics or chemistry, nor is it necessary to memorize comprehensive factual information about the scientific basis of our modern technological civilization. If one understands the nature of modern science, such factual information can when needed be readily acquired by consulting appropriate books and articles.

A direct study of the methods of modern science presents great difficulties. A visitor to a laboratory, unless he is himself a scientist, will find it almost impossible to understand the work in progress; he will comprehend neither the objectives nor the implications of the measurements and observations that the investigator is making. To be sure, he may be able to appreciate in a general way the nature of the problem that is under investigation; he may well realize that the questions raised by those who planned the experiment are of importance — either for immediate practical reasons or as part of the general advance of science. But unless he himself has had considerable experience with scientific research, he will be baffled by what he sees or hears, for almost without exception research problems today are concerned with areas of science where there is a large accumulated background of technical information. To be able to follow the new work one must have studied the advance of the science thoroughly, which usually means mastering a considerable amount of mathematics as well as physics or chemistry or biology.

Modern science has become so complicated that today methods of research cannot be studied by looking over the shoulder of the scientists at work. If one could transport a visitor, however, to a laboratory where significant results were being obtained at an early stage in the history of a particular science, the situation would be far different. For when a science is in its infancy, and a new field is opened by a great pioneer, the relevant information of the past can be summed up in a relatively brief compass. Indeed, if the methods of experimental science are being successfully applied for the first time to a problem of importance, the scientist's knowledge would not be much greater than that of his inquiring guest. Briefly, and in simple language, he could explain the new experiment. Then as from day to day results were obtained and further experiments planned the visitor would see unfolding before him a new field. However, such periods in the history of science are relatively few, and it would be a fortunate visitor indeed who could spend some time merely as a witness of the events. For he would have the pleasure of knowing not only that he had been present at one of the critical battles in the forward march of science, but that he had had a unique opportunity of learning at first hand about the methods of science.

2. THE USE OF CASE HISTORIES

The purpose of the case histories presented in this series is to assist the student in recapturing the experience of those who once participated in exciting events in scientific history. The study of a case may be to some degree the equivalent of the magical operation suggested in the preceding paragraph, namely, that of transporting an uninformed layman to the scene of a revolutionary advance in science. To be sure, usually more than one investigator or laboratory is involved and the period of time is at least several years; for, however significant a single experiment may seem in retrospect, no important step forward in experimental science rests solely on the record of any single investigator's observations. Rather, the interplay of ideas of several men, arguments about experiments and their interpretation between workers in different laboratories, often mark the decisive turn in scientific thinking. However, by the intensive study of the actual work of some of the great investigators of the past (and in less detail, the work of their contemporaries), the methods then employed stand out clearly.

Some of the cases in this series present the work of men who lived over three hundred years ago; others are drawn from the eighteenth and nineteenth centuries; a few may involve twentieth-century discoveries. But irrespective of their dates, the examples presented illustrate the methods of modern science. Familiarity with those methods will increase the ease of understanding of the work of scientists today; as one consequence, popular accounts of scientific progress can be read with more appreciation. If one is face to face with problems of financing or organizing research, a critical but sympathetic attitude toward experimental investigation is a prerequisite for an intelligent examination. The occasion may arise in connection with private industry, the expenditure of public money, or a philanthropic enterprise related to medicine and public health. Almost all that a trained scientist has to go on when he passes judgment on the prospects of a new venture far removed from his own specialty is his knowledge of the methods by which science has progressed in his own experience. To a considerable degree a nonscientist may come to have a similar basis for his opinions. An intimate acquaintance with a relatively few historic cases should assist him in finding his way through the complexities of modern investigation as he listens to those who tell him what is being proposed.

3. CONCERNING THE METHODS OF SCIENCE

Science we shall define as a series of concepts or conceptual schemes arising out of experiment and observation and leading to new experiments and new observations. From the experimental work and careful observations of nature come the scientific facts that are tied together by the concepts and conceptual schemes of modern science. The rather sudden burst of interest in the new "experimental philosophy" in the seventeenth century is historically related to a new interest on the part of thoughtful men in practical matters ranging from medicine through mining to the ballistics of cannon balls. But while the problems were often suggested by the interest of learned men in the practical arts, the development of science involved something more than the type of experimentation by which the practical arts had been developing for centuries. The method of reasoning employed in mathematics (for example, in geometry), commonly called deductive reasoning, had to be combined with the methods of experimentation that came from the practical arts before science could progress rapidly. The interaction of these two streams of human activity was largely responsible for the development of physics and chemistry; the focus of attention was shifted from an immediate task of improving a machine or a process to a curiosity about the phenomena in question. New ideas or concepts began to be as important as new inventions, and their interweaving began to produce conceptual fabrics whose various threads gained support from one another as well as from the direct results of experiment and observation.

Concepts and Conceptual Schemes. We have defined science as a series of concepts or conceptual schemes (theories) arising out of experiment or observation and leading to new experiments and observations. The cases presented in this series illustrate some of the many ways in which the new ideas (concepts) have arisen and the consequences they have entailed. In more than one case we shall see a new conceptual scheme (theory) replacing an older one, or two rival schemes in conflict. The transition to a new theory is seldom easy; old ideas are apt to be tenacious. Looking back at the history of any branch of science we can see that a new conceptual scheme (theory) comes to be accepted because it is at least as satisfactory as the older one in relating all the known facts in a simple way and because it proves to be more fruitful of new experiments. The latter outcome is clearly a matter for the future, so that there can be no way to determine in advance whether a new concept or conceptual scheme will prove acceptable in this double sense; nor can one foretell how soon the next major advance will occur. A study of these cases makes it clear that there is no such thing as *the* scientific method, that there is no single type of conceptual scheme and no set of rules specifying how the next advance will be made through the jungle of facts that are presented by the practical arts on the one hand and by the experimentation and observation of scientists on the other.

Speculative Ideas and Broad Working Hypotheses. The relation between a speculative idea and a broad working hypothesis that becomes a wide conceptual scheme is of interest. A good example is furnished by the history of the atomic theory. The notion that there are fundamental units — ultimate particles — of which matter is composed goes back to ancient times. But expressed merely in general terms this is a speculative idea until it forms the basis of a broad working hypothesis from which consequences can be deduced that are capable of experimental test. This particular speculative idea became in Dalton's hands a broad working hypothesis and came to be a new conceptual scheme only after he had shown, about 1800, how fruitful it was in connection with the quantitative chemical experimentation that had been initiated by the chemical revolution.

Dalton's atomic theory is an instance where we can trace the origin of a conceptual scheme; in other cases we are uncertain how the idea came to the proponent's mind. Thus Torricelli seems to have been the first to suggest that we live in a "sea of air" that exerts a pressure analogous in many ways to the pressure of water on submerged objects; yet we have insufficient knowledge to say what led him to this idea. His famous experiment with a column of mercury, in which he made the first barometer, may have been the point of departure or it may have been a case of testing a deduction from a broad working hypothesis, arrived at by means that history does not record. At all events, from that time on a number of deduced consequences led to experiments nearly all of which were consistent with the ideas of Torricelli, and his hypothesis quickly attained the status of a new conceptual scheme.

The cases presented in this series emphasize the prime importance of a broad working hypothesis that eventually becomes a new conceptual scheme. They illustrate the variety of mental processes by which the pioneers of science developed their new ideas. But it must be emphasized that great working hypotheses have in the past often originated in the minds of these scientific pioneers as a result of mental processes that can best be described by such words as "inspired guess," "intuitive hunch," or "brilliant flash of imagination." The origins of the working hypotheses are to be found almost without exception in previous speculative ideas or in the previously known observations or experimental results. Only rarely, however, do these broad working hypotheses seem to have been the product of a careful examination of all the facts and a logical analysis of various ways of formulating a new principle.

The Testing of Deductions from Conceptual Schemes. When first put forward, a new conceptual scheme may be no more than a working hypothesis on a grand scale. From it we can deduce, however, many *consequences*, each of which can be tested by experiment. If these tests confirm the deductions in a number of instances, the accumulating evidence tends to confirm the broad working hypothesis and the hypothesis may soon come to be accepted as a new conceptual scheme; its subsequent life may be short or long, for from it new consequences are constantly being deduced that can be verified or not by experimental test.

While the modes of formulating broad working hypotheses are so varied as to defy generalization, the procedures by which these hypotheses are tested conform to a fairly consistent pattern. One is tempted, therefore, to represent the process of testing a broad conceptual scheme by a diagram. Unfortunately, however, the interrelations are so complex as to be more confusing than helpful. One cannot grasp an entire case at a glance; to get the real point of the story it must be studied in a step-by-step fashion at a reasonable pace. These Case Histories in Experimental Science provide the material for just such study.

While our purpose would be defeated in the distortion necessary to compress a case history into a single diagram, we can perhaps suggest by a particular example, drawn from *part* of one case, the type of process involved in the experimental testing of broad conceptual schemes. Thus, at one stage in the development of pneumatics, Pascal deduced from Torricelli's idea of a sea of air the consequence that the air pressure should be less on a mountain top than at the base. This deduction could be expressed in some such words as these: if the Torricellian experiment is performed at such and such a place the height of the mercury column will be greater than on the same day at a nearby spot high up on a mountain. This prediction is susceptible of a test which yields a yes or no answer.

As the cases illustrate, however, there are often many slips in the argument at this point; unconscious assumptions are often involved that turn out to have been unwarranted. In general, the complete pattern of deduction linking the broad working hypotheses to the experimental tests proves far from simple. Thus, in the example just cited, the tacit assumption is clearly being made that the laws of liquid pressure and the density of mercury (and probably several other such things as well) are not themselves appreciably different at different altitudes; these assumptions find their ultimate justification (for the purposes of *this* experiment, at least) in other broad conceptual schemes such as make up the laws of hydrostatics. It is the multiplicity, the complexity, and indeed often the vagueness of this sort of interconnection that is ignored in many oversimplified discussions of "*the* scientific method."

Experimentation. The experiments performed to test deductions from broad working hypotheses involve in the last analysis a type of activity with which everyone is familiar. Examples of experimentation in everyday life come readily to mind. Whenever one has a choice of alternative ways of trying to solve a problem, decides on one, and then says in essence "let's try it and see," he is getting ready to perform an experiment. Framing various possibilities as to what is wrong with an automobile engine when the car is stalled is usually a preliminary to carrying out an actual trial, an experiment. Similarly, if a man is trying to locate a broken wire, a blown-out fuse, or a bad tube in a radio set, he usually turns over in his mind a number of tests that could be made and then tries first one and then another. He is guided in the formulation of his trials (experiments) by his knowledge of the automobile or the radio set in question and his general information about the behavior of these machines. When he finally carries out the experiment he is, however, testing the validity of a very *limited working hypothesis.* This limited working hypothesis can generally be stated in some such form as "if I do so and so, then such and such will happen." A simple and perhaps trivial example of the testing of a limited working hypothesis is when a person trying to unlock a door puts in the lock a hitherto untried key and says to himself, "if I turn the key, then the lock will spring." The trial either validates or negates the limited working hypothesis; the same is true of the final step in most experiments in science.

The difference between experimentation in everyday life and experimentation in science lies primarily in the fact that in the one case common-sense assumptions and practical experience determine the nature of the experiment; in the other, a series of connecting links usually relates some deductions of a broad working hypothesis with the final limited working hypothesis involved in the specific experiment. This will be more evident as one studies the cases. The steps by which practical men throughout the ages have improved the practical arts have also involved experimentation. Here, as in everyday life, however, the experiment is in almost every case planned in terms of common-sense assumptions and practical experience, not in terms of broad generalizations or conceptual schemes. Furthermore, as was pointed out in the opening paragraph of this section, experimentation in the practical arts always has a practical objective. Experimental science was the consequence of the combination of the ancient art of experimentation with the type of thinking that employs general ideas and deductive reasoning. Experimentation by itself does not produce science, although it is an essential element in advance in many fields of science.

The progress of science in many areas has depended on the overcoming of experimental difficulties, in part by the invention of new instruments and the discovery of new materials. A study of the types of difficulties that have arisen in the past and how they have interfered with the testing of new concepts and conceptual schemes throws light on what is going on in the laboratories of physics and chemistry today. Furthermore, in formulating problems according to certain conceptual schemes, and particularly in planning the experimental tests, it became necessary as science advanced to make precise and accurate many vague common-sense ideas, notably those connected with measurement. Old ideas were clarified or new ones introduced. These are the new concepts that are often quite as important as the broad conceptual schemes; for example, pressure, acceleration, mass, temperature, atom, are words that have now precise scientific meanings. At the time the new ideas were advanced that are considered in the case histories from the seventeenth, eighteenth, and nineteenth centuries, they were as novel as are in our own times the ideas of neutrinos and mesons in the field of nuclear physics.

A close attention to the difficulties that have beset investigators in getting clean-cut answers to apparently simple questions will suggest some of the complexities of research today. And the broader hypotheses must remain speculative ideas until one can by the processes indicated succeed in relating them to experiments.

4. THE ADVANCE OF SCIENCE AND PROGRESS IN THE PRACTICAL ARTS

The relation between advances in the practical arts (such as mining, agriculture, manufacturing) and progress in science is more complicated than at first sight appears. To appreciate the present situation, it is helpful to have some knowledge of the history of these two branches of human endeavor and of their interaction. For many generations after the new experimental philosophy was put forward by a few amateurs in the seventeenth century, the direct impact of science on the practical arts was almost negligible. This was the consequence not of any lack of interest in practical affairs on the part of the early scientists but of the fragmentary nature of the knowledge they acquired and the inadequacy of the conceptual schemes they elaborated. The advances in the practical arts of mining, metal making, agriculture, food processing, and medicine in the eighteenth and early nineteenth centuries were made without any very direct benefit of science. (The art of navigation is an exception to this statement.)

The ancient method of experiment, the use of the "let's-try-it-and-see" type of reasoning in solving immediate practical problems, has led over the centuries to amazing results. This process of advance is often called *pure empiricism*, there being no wide generalization to guide the inventor. Even today in the mid-twentieth century when science has penetrated so deeply into industry, there is still a great deal of empiricism in a practical art such as steel making. Even when no practical aim is involved, the practical methods of a science may contain many recipes and rules that have no relation to theory; in other words, there is some empiricism in almost all scientific procedures even now. It is often convenient to speak of the "degree of empiricism" in a given industrial process or branch of science. Where wide generalizations and theory enable one to calculate in advance the results of an experiment or to design a machine (a microscope or a telescope, for example), we may say that the degree of empiricism is low. On the other hand, if the conditions for satisfactory operation of a machine, a process, or an instrument are based only on cut-and-try experimentation (as is true in some branches of chemistry), we may say that the degree of empiricism in that branch of science is still high.

5. PURE AND APPLIED SCIENCE

Applied research may be defined as research directed to the end of reducing the degree of empiricism in a practical art. Activities concerned solely with developing and improving our conceptual schemes and filling in gaps in our systematic knowledge may be defined as pure science. Modern science is a fabric the texture of which is composed of many interrelated concepts and conceptual schemes. Today the validity of each conceptual scheme must be tested in the light of our total knowledge. The cases presented in this series fall in the category of pure science; they are nevertheless concerned for the most part with episodes in the history of science that eventually had great influence on applied science. The conceptual schemes elaborated by scientists have been fundamental to further discoveries and to the formulation of the ideas that are the fabric of modern science. With the aid of the concepts and theories developed over the past three hundred years, and drawing on the systematic knowledge accumulated during that period, the applied scientist today can improve the industrial arts and invent new processes, products, and machines far more readily than could his predecessors of the seventeenth and eighteenth centuries. Science today is closely connected with technology because theory and the practical arts have become closely interwoven. The degree of empiricism has been lowered on almost every hand and is being still further reduced by the activity of investigators in laboratories of both pure and applied science. The applied scientist or engineer can now engage in what was once an activity of those inventors who proceeded largely empirically. The application of science to new industrial processes is often designated "development work"; applied research and development today go hand in hand.

The analysis of experimental science presented in this foreword and a study of the cases outlined in this series will, it is hoped, lead the layman to a better understanding both of modern science and of modern technology. No college course can be a substitute for experience; it can only lay a foundation. With an introduction to an understanding of experimental science, the inquiring citizen ought to be in a position to continue his scientific education throughout his life. In the belief that a detailed knowledge of a few epoch-making advances in science will provide a key to a better comprehension of the modern world, the Harvard Case Histories in Experimental Science are presented to the public.

JAMES B. CONANT

Cambridge, Massachusetts
November 1, 1951

CASE 7

Pasteur's and Tyndall's Study of Spontaneous Generation

GENERAL INTRODUCTION

Louis Pasteur (1822–1895) was a chemist whose studies led him to investigate the biological process known as fermentation (Case 6). Early in these studies Pasteur put forward the hypothesis that fermentation is a process associated with the growth of living organisms and not with their death and decay. Having taken this position, he was impelled to enter into the ancient controversy regarding the possibility of spontaneous generation of living organisms from inert matter. His first comprehensive paper dealing with the subject was published in 1862 under the title "Memoir on the Organized Corpuscles that Exist in the Atmosphere; A Study of the Doctrine of Spontaneous Generation." It is a lengthy document. In the first section Pasteur recounts the history of the subject and explains how he came to take up an inquiry which till then "had taxed the skill and wisdom of naturalists alone." A translation of this portion of the "Memoir" constitutes Section 1 of the present case.

Pasteur refers briefly to the experiments of Francesco Redi (1626–1697). Section 2 of this case consists of a translation of part of Redi's *Experiments on the Generation of Insects*. This book, written in Italian, was first published in 1668 and in 1688 reached the fifth edition. Redi studied medicine at the University of Pisa and was court physician to Ferdinand Medici, Grand Duke of Tuscany. Pasteur describes Redi as "a celebrated member of the Accademia del Cimento." This, the Academy of Experiments, was organized in 1657 in Florence by a group of former students of Galileo who had been meeting together since 1651. When the patron of this group, Leopold Medici, became a cardinal in 1667, the Academy dissolved. The contributions of the members of the Accademia del Cimento to our knowledge of pneumatics are referred to in Case 1 (footnote, p. 13).

Redi's account of his experiments illustrates how old is the art of experimentation (see pp. 4–8 of the Foreword). Even the "control" so frequently employed in biological experimentation today is part of

Redi's technique (Section 2, p. 29). Yet Redi makes no claim to any new method of studying nature. He uses almost as a matter of course a trial-and-error procedure. Such ways of obtaining answers to highly limited working hypotheses (Foreword, p. 7) are as old as the development of the practical arts. Redi's working hypothesis is not a very broad one, though it is not related to a practical end. He is concerned solely with "the generation of insects" and demonstrates in the sections here reprinted only that flies are the necessary precursors of the worms ordinarily found in certain putrefying meats. The difficulties of generalizing from one set of experimental data to all other instances of apparent spontaneous generation are illustrated in the other sections of this case.

Sections 3, 4, and 5 are translations of parts of the exhaustive report of Pasteur's experiments presented in his 1862 Memoir. Section 6 is from a more popular exposition, by the English physicist John Tyndall (1820–1893), of the sort of evidence that has convinced most people of the errors of those who held to the doctrine of spontaneous generation. The student would do well to read in this same connection Tyndall's lecture on *Fermentation, and Its Bearing on Surgery and Medicine* (Case 6, pp. 36–57). In reading these papers by Pasteur and Tyndall one must bear in mind the significant advance that had been made by Pasteur when he published his *Memoir on Lactic Fermentation* (Case 6) in 1858. By 1862, when Pasteur first published his views on spontaneous generation, he had studied alcoholic fermentation in some detail. The new facts which he had brought to light were, he believed, additional evidence for the correctness of his hypothesis that alcoholic fermentation is "correlative to life and to the organization of globules [of yeast], and not to their death or putrefaction" (Case 6, p. 32).

Pasteur had also shown that the spores of the mold *Penicillium glaucum* would develop in the absence of air in a medium containing optically inactive tartaric acid—that is, a tartaric acid solution that would rotate a beam of plane-polarized light neither to the right nor to the left (Case 6, pp. 22–23). The apparent lack of optical activity was thought to be due to the presence in the tartaric acid of exactly equal proportions of two optically active isomers: one rotating the plane of polarization to the right, the other to the left. After the development of the mold the tartaric acid that remained was found to be optically active, rotating to the left. This suggested that the molecules which rotated the plane of polarized light to the right were selectively used up; those that rotated to the left were either not consumed or consumed at a slower rate. Pasteur had also isolated the organism responsible for butyric acid fermentation. In short, he had laid the foundations for experimental microbiology, of which bacteriology was soon to become a most important subdivision.

With the techniques thus available to him, Pasteur was able to study the alleged cases of spontaneous generation with great effectiveness. Indeed, in 1864 he was able to convince a commission of the French Academy of the correctness of the thesis that he had put forward in Chapter VII of his large memoir on spontaneous generation, the thesis: "It is not true that the smallest quantity of ordinary air is sufficient to produce in an infusion organized life characteristic of that infusion." (An infusion is a water extract of material of animal or vegetable origin. Chapter VII of Pasteur's memoir is reproduced in part in Sec. 4 of this case.) The commission of the French Academy had been appointed to adjudicate the dispute that had arisen between Pasteur and the French naturalist Félix A. Pouchet (1800–1872), who maintained an opinion diametrically opposed to that expressed by Pasteur in the last quotation.

Pouchet had carried out experiments very similar to those described by Pasteur. He too prepared flasks containing fermentable material, sterilized them and their contents by boiling (during which the air originally in the flasks was expelled), sealed off the necks, and observed no change as long as the flasks were unopened. But when he opened the flasks, even on the edge of a glacier at an elevation of 6000 feet, admitting air and then resealing the vessels, *all* the flasks were subsequently found to contain growing organisms. Thus the admission of the pure air at the edge of the glacier had, apparently, sufficed to produce a spontaneous generation of living organisms in the nutrient medium. Pasteur's experimental results were quite different. For example, of 19 flasks opened and then sealed in the lecture hall of the Museum of Natural History, only 4 showed growths; of 18 opened outdoors under some trees, 16 developed growths. Thus Pasteur's results indicated that the admission of air did not absolutely ensure the subsequent development in the flasks of living organisms. From this he inferred that these growths were produced not by the air itself, but by minute seeds or spores that might or might not be present in any small specimen of air. For some reason Pouchet and his collaborators raised trivial objections to Pasteur's experiments and withdrew from the competition arranged by the French Academy's commission.

But subsequent history showed that both Pasteur and Pouchet were right in so far as the experiments were concerned. Pouchet had not failed, as Pasteur thought, by careless technique (which, of course, very often yields results that appear to be the consequences of spontaneous generation). One of Pasteur's young assistants, Duclaux, writing of these days long afterward, said: "The battle was won, for Pasteur was sure of his experiments . . . Had anyone told us that this brilliant victory amounted to nothing, he would have surprised us very much. Nevertheless, such was the case. Pasteur was right; Pouchet [and his collaborators], Joly and Musset were right also, and if, instead of withdrawing, they had repeated their experiments, they would have embarrassed the Commission very much, and Pasteur would not then have known how to reply to them.

"It is in reality quite true that if one opens, at any point whatsoever on the globe, flasks filled with a *decoction of hay*, as Pouchet did, it often happens that *all* the flasks become clouded and filled with living organisms. In other words, with this infusion [of hay] the experiments of Pasteur with *yeast water* do not succeed . . . The fact is that the germs already exist in the [hay] infusion. They have resisted boiling, as is the case with a great number of microörganisms. They have remained inert as long as the flask, sealed during the boiling, remains devoid of air. They develop when the air enters, thanks to its oxygen. But Pasteur did not yet know this result."

As a matter of fact, it was only in 1876, as a result of a controversy with an English doctor, Henry Charlton Bastian (1837–1915), that Pasteur became aware of the resistance of the spores of certain bacteria to long exposure to the temperature of boiling water. The spores of the bacilli in hay are not only unusually resistant but able to *develop only in the presence of oxygen*. John Tyndall was also involved in the controversy with Dr. Bastian, and the last section of this case is a popular account of Tyndall's experiments which showed the resistant nature of the hay-germs.

Three points of special interest stand out from this historic mistake of Pasteur's in the 1860's. The first is how easy it is to *assume* that two sets of experimental conditions are effectively identical in spite of their obvious differences. Why did not Pasteur try Pouchet's experiments with hay infusion? Probably because he was convinced that all fermentable materials were *essentially* the same (yeast water with sugar, vegetable infusions, mutton broth, what did it matter?).

The second point to be noted is the practical consequences of the demonstration that the spores of certain bacteria are heat-resistant. There soon developed the practice of sterilizing at temperatures above the boiling point of water, by using an autoclave (that is, a modified pressure cooker). This is routine procedure in bacteriology today.

The third point is more philosophical in nature. Every day, probably, someone in some laboratory or hospital finds a supposedly sterile medium that shows growths. But no one in his or her right mind would suggest that this was due to spontaneous generation. Rather, it would be assumed that some error had been made, that the flasks or media had not been sufficiently sterilized. And no one today would take the time and trouble to study further such cases of apparent spontaneous generation. The impossibility of the kind of spontaneous generation that Pouchet and Bastian believed in has thus become an accepted premise among scientists. Why is this so? Are Pasteur's and Tyndall's experiments so convincing? Can one safely generalize so widely even from a mass of negative evidence, particularly if one must, apparently arbitrarily, throw out the positive cases as being due to "faulty manipulation?"

The ghost of Needham (see p. 19) might even rise to protest against the use in this work of the autoclave and the repeated sterilizations which we say are necessary to destroy heat-resistant bacterial spores. "You have destroyed the vegetative force," he might declare, "you have so tortured the proteins (if he used modern language) that the molecules will no longer come together spontaneously as organized globules, as living entities." The answer a modern bacteriologist would probably give represents the basic reason why no one today believes in spontaneous generation. Thanks to the techniques first suggested by Pasteur in his study of lactic acid fermentation, it is now possible to obtain a *pure* culture of a specific microörganism. This art has so far developed that one no longer has to work with accidental mixtures of microörganisms as did Pasteur, Pouchet, Bastian, and Tyndall (excepting Tyndall's work with hay-germs). Now, the striking fact is that after inoculation with a given microörganism a sterile medium shows the growth of the *same* organism. Needham's ghost would have to explain why the "vegetative force" in a given medium yielded bacteria A when inoculated with a trace of A and bacteria B when inoculated with a trace of B. The most convincing evidence for Pasteur's thesis of the impossibility of spontaneous generation is, therefore, to be found in the whole fabric of the results of the study of *pure* cultures in the last sixty or seventy years. But without the pioneer work recorded in the present case, none of this would have been possible. As Duclaux remarks in summing up the Pouchet-Pasteur story: "This is a good illustration of what a series of judgments, revised without ceasing, goes to make up the incontestable progress of science. We must believe in this progress, but we must never accord more than a limited amount of confidence to the forms in which it is successively vested. One sometimes reaches the truth by error, and sometimes the error by truth." [Duclaux, *Pasteur; The History of a Mind* (Philadelphia and London, 1920), p. 111.]

1. TRANSLATION OF A PORTION OF PASTEUR'S "MEMOIR ON THE ORGANIZED CORPUSCLES WHICH EXIST IN THE ATMOSPHERE"

The principal results described in this "Memoir" were presented to the French Academy of Sciences at its meetings of February 6, May 7, September 3, and November 12, 1860. The "Memoir" was published in *Annales de Chimie et de Physique* (3rd series) **64**, 5–110 (1862). The material in square brackets has been added by the editor of this case.

CHAPTER I

HISTORICAL[1]

From ancient times until the end of the Middle Ages everyone believed in the existence of spontaneous generation. Aristotle says that all dry things which become moist and all moist things which become dry engender animals. Van Helmont [the Belgian physician and chemist, J. B. van Helmont (1577–1644)] describes the way to produce mice. Even in the seventeenth century many authors describe ways of bringing forth frogs from the mud of marshes or eels from the water of our rivers. Such errors could not, however, long survive the spirit of inquiry which took hold of Europe in the sixteenth and seventeenth centuries. Redi, a celebrated member of the *Accademia del Cimento*, demonstrated that the worms in putrefying flesh were larvae from the eggs of flies. His proofs were as simple as they were decisive, for he showed that if the putrefying flesh were surrounded with fine gauze the hatching of these larvae was absolutely prevented.

Redi was also the first to recognize the males, the females, and the eggs of those animals that live in other animals. Réaumur [the naturalist René A. F. de Réaumur (1683–1757) displayed his versatility in a number of different fields of pure and applied research] later said that one could surprise these flies in the act of depositing their eggs in fruits: when one saw a worm in an apple one knew that it was not decay which engendered the worm but, on the contrary, that the worm was the cause of the rotting of the fruit.

But soon, in the second part of the seventeenth century and the first half of the eighteenth, microscopic observations rapidly increased in number. The doctrine of spontaneous generation then reappeared. Some investigators, unable to explain the origin of the varied organisms which the microscope showed in infusions prepared from animal or vegetable materials, and seeing among these organisms nothing that resembled a process of sexual generation, were led to assume that matter once alive keeps after death a special vitality. It was further assumed that under the influence of this vitality the scattered parts unite once more, under certain favorable conditions, with variations of structure and organization determined by these same conditions.

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[1] The reader will note that one of my chief concerns in presenting this historical material has been to assign proper credit to each experimenter. But I have taken equal care not to confuse true progress either with the numerous essays which have appeared on the subject or with ambiguous experiments which hamper rather than assist the progress of science. In this sort of question, which has been considered for centuries by so many thinkers, all manner of a priori views, all arguments by analogy or by induction, and all hypotheses have come to light. What is truly important is rigorous proof; that is, the conduct of experiments *freed from all the confusions which normally arise from the experiments themselves.*

Other investigators, on the contrary, using their imaginations to supplement the marvelous discoveries made with the microscope, believed they saw couplings among these Infusoria, and males, females and eggs: they therefore declared themselves as open adversaries of the doctrine of spontaneous generation. One must admit that the proofs used to support either of these opinions did not bear examination.

The question was still open when in 1745 there appeared in London a work by Needham [John T. Needham (1713–1781)], skillful observer and Catholic priest of ardent faith — a circumstance which in such a matter was equivalent to a guarantee of the sincerity of his convictions. The doctrine of spontaneous generation was supported in this work by facts of an entirely new order. I refer to the experiments with hermatically sealed vessels that had previously been exposed to an elevated temperature. Indeed, it was Needham who first conceived the possibility of such experiments. Less than two years after the publication of Needham's results the Royal Society of London elected him to membership. Later he became one of the eight [foreign] associates of the Académie des Sciences. But, above all, Needham's work became famous because of the support that it received from Buffon's systematic treatment of the subject of generation. [George L. Leclerc, Comte de Buffon (1707–1788), was an enormously influential encyclopedist of the scientific knowledge of his time. Fifty-five years in preparation, the 44 volumes of Buffon's treatise were completed after his death by an assistant.]

The first three volumes of Buffon, in the quarto edition published during his lifetime, appeared in 1749. In the second volume of this edition, four years after the appearance of Needham's book, Buffon expounded his system of organic molecules and defended the hypothesis of spontaneous generation. Presumably Needham's results had a great influence on Buffon's views, for in the same period when the illustrious naturalist was preparing the first volumes of his work, Needham made a trip to Paris during which he became the guest of Buffon and to some extent his collaborator.

The ideas of Needham and Buffon had both partisans and detractors. These ideas were opposed by another famous doctrine, that of Bonnet [Charles Bonnet (1720–1793)], who assumed the preëxistence of germs. The contest was the more lively since each side was convinced of the truth of its own opinions. We know today that the truth was neither on the one side nor on the other. But we must remember that these were the days when men were eager to enter exhaustive discussions of rival systems and speculative views.

There were, we may say, two contradictory persons in Buffon himself. One stated explicitly that he sought for a hypothesis in order to erect a system, and the other wrote for his translation of the *Vegetable Staticks* by Hales [Stephen Hales (1677–1761)] the fine preface in which the necessity for experiment was given a deservedly high place. These two sides of the genius of Buffon were to be found in different degrees in all the learned men of his period. But Needham's conclusions were soon to be submitted to experimental verification. There was at that time in Italy a physiologist, one of the most skillful who ever adorned science, and one of the most ingenious and most difficult to satisfy as well — the Abbé Spallanzani [Lazaro Spallanzani (1729–1799)].

Needham, as I have just said, had supported the doctrine of spontaneous generation by direct experiments which were very well conceived. Only experiment could serve either to overthrow or to reinforce his opinions. This Spallanzani understood very well. "In many cities in Italy," he says, "one sees men opposing the opinion of Needham; but I do not believe that anyone has ever thought of examining it experimentally."

In 1765 Spallanzani published at Modena a dissertation in which he refuted the ideas of Needham and Buffon. This work was translated into French, probably at the request of Needham, for the edition which appeared in 1769 carried notes written by him in which he answered all of Spallanzani's objections. The latter, struck no doubt by the justice of Needham's criticisms, returned once more to his work and soon produced the fine collection of studies detailed in his *Opuscules physiques*.

It would be useless to present a complete history of the dispute between these two learned naturalists. But it is important to note precisely the experimental difficulty at which their efforts were particularly directed, and to find out whether this long controversy really banished all doubt, as is generally believed. Spallanzani is usually regarded as the victorious adversary of Needham. If this judgment were well founded, is it not astonishing that there should still be in our day so many partisans of the doctrine of spontaneous generation? In science does not error usually disappear much more promptly, even in questions of this sort, when it has really been unmasked by experiment? Is it not to be feared that when error is reborn in good faith its defeat was only apparent? An impartial examination of the contradictory observations of Spallanzani and Needham on the critical point at issue shows us that, contrary to the generally accepted opinion, Needham was not really forced to abandon his doctrine because of Spallanzani's work.

I have said that Needham was the originator of experiments dealing with what can be observed in sealed vessels previously exposed to the action of heat.

"We are assured by Needham," says Spallanzani, "that experiments thus carried out have always been highly successful in his hands, that is to say that the infusions have shown Infusoria; and that it is this result which puts the seal [of approval] on his system."

"If," adds Spallanzani, "after having purged by means of heat both the substances in the vessels and the air contained in these same vessels, one takes the further precaution of cutting them off from all communication with the surrounding air; and if, nevertheless, on opening the vessels one still finds living animals, this would be strong evidence against the ovarian system: *I scarcely know how its partisans could reply to this.*"

I underline these last words in order to show that Spallanzani considered the criterion of truth or error to lie in the results of experiments thus conducted. But the following quotation from Needham's notes shows us that this was equally his opinion. Here, indeed, is a passage from Needham's remarks on Chapter X of Spallanzani's first treatise.

"Nothing remains to be done," says Needham, "save to speak of Spallanzani's last experiment, which he himself believes to be the only one in his entire treatise that appears to have some force against my ideas. He hermetically sealed nineteen vessels filled with different vegetable substances and he boiled them, thus closed, for the period of an hour. But from the method of treatment by which he has tortured his nineteen vegetable infusions, it is plain that he has greatly weakened, or perhaps entirely destroyed, the *vegetative force* of the infused substances. And, not only this, he has, by the exhalations [from the vegetables] and by the intensity of the fire, entirely spoiled the small amount of air that remained in the empty part of his vessels. Consequently, it is not surprising that his infusions, thus treated, gave no sign of life. This is as it should have been.

"Here then, in a few words, is my last proposition and the result of all my work: Let him renew his experiments, using substances sufficiently cooked to destroy all the supposed germs that one may believe to be attached to the substances themselves or to the interior walls of the vessel, or floating in the air within the vessel. Let him seal his vessels hermetically, leaving within them a certain amount of undisturbed air. Let him then plunge the vessels into boiling water for several minutes, the time which is necessary to harden a hen's egg and to kill the germs. In a word, let him take all the precautions that he wishes, provided that he seek only to destroy the supposed foreign germs which come from the outside. And I reply that he will always find these microscopic living creatures in number sufficient to prove the correctness of my ideas. If, having conformed with these conditions, he finds on opening his vessels (after having left them quietly for the time necessary for the generation of these organisms) nothing alive nor any sign of life, then I abandon my system and renounce my ideas. This is, I believe, all that a reasonable adversary can ask of me."

Here certainly the issue between Needham and Spallanzani is clearly drawn. In the third chapter of the first volume of his *Opuscules* Spallanzani tackles the decisive difficulty. And what is his conclusion? In order to suppress all production of Infusoria it is necessary to keep the infusions for three-quarters of an hour at the temperature of boiling water. But does not the necessity of an exposure of three-quarters of an hour to a temperature of 100 degrees justify Needham's fears about a possible alteration of the air in the vessels? At the very least, Spallanzani should have added to his experiments an analysis of this air. But science was not yet sufficiently advanced: eudiometry [methods for the analysis of gases and, notably, for the determination of the oxygen content of air] had not yet been created. The composition of atmospheric air was scarcely known.[1]

The results of Spallanzani's experiments on the crucial point in the whole question thus remained open to the full force of Needham's objections. Moreover these objections were, at least in appearance, validated by the subsequent progress of science. Appert applied to domestic economy the results of Spallanzani's experiments carried out according to the method of Needham. [In 1810 the French investigator Appert published an account of his invention of the process we now call "canning."] For example, one of Spallanzani's experiments consisted in putting green peas with water into a glass vessel which was later hermetically closed, after which it was placed in boiling water for three-quarters of an hour. This was also the procedure of Appert. But Gay-Lussac [J. L. Gay-Lussac (1778–1850), an eminent French chemist], desiring to investigate this procedure, submitted it to diverse trials, the results of which he published in one of the most frequently quoted of his Memoirs.

The following extracts from the work of Gay-Lussac leave no doubt about the opinion of this illustrious natural philosopher, an opinion which has passed into science complete and uncontested:

"Analysis makes it evident," says Gay-Lussac, "that there is no oxygen left in the air of bottles in which such substances as beef, mutton, fish, mushrooms and must of grapes [grape juice prepared for wine-making] have been satisfactorily preserved; *and, consequently, that the absence of oxygen is a necessary condition for the preservation of animal and vegetable substances*." [In a footnote Pasteur comments on the logical inadequacy of Gay-Lussac's conclusion.]

Needham's fears that there had been an alteration in the air in the vessels used in Spallanzani's experiments were thus justified by the fact that oxygen was absent from Appert's preserves. But notable progress was soon made as a result of an experiment by Dr. Schwann [the German physician Theodor Schwann (1810–1882)]. In the month of February 1837, Schwann reported the following facts: An infusion of lean meat is placed in a glass flask; the flask is then sealed off in a flame, and the entire system is exposed to the temperature of boiling water. Left to itself, after cooling, the liquid does not putrefy. So far nothing new had been discovered. This was one of Spallanzani's experiments or, one might better say, the preparation of one of Appert's preserves. But it was desirable, adds Schwann, to modify the experiment in such a way that a renewal of the air in the flask becomes possible, under conditions which insure that the incoming air is first heated, as was the air originally in the flask. To this end Schwann repeated the preceding experiment with a flask to whose neck had been fitted a two-holed stopper through which passed glass tubes. These tubes were bent so that a section of each could be immersed in a bath of fusible alloy maintained at a temperature close to that of boiling mercury. The air is renewed by means of an aspirator; the air comes into the flask cool, but this air has already been heated during its passage through the part of the tubes surrounded by the fusible alloy. One begins the experiment by boiling the liquid [and the rest of the experiment follows as before]. The result is the same as in the experiments of Spallanzani and Appert. There is no alteration of the organic liquid.

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[1] Spallanzani's first treatise appeared in 1763. His *Opuscules* appeared for the first time in 1776. Lavoisier's discovery of the composition of air was made in 1774.

Air that has been heated and then cooled does not, therefore, cause an alteration in meat juice which has been brought to the boiling point. This was a great step forward, because it reinforced Spallanzani's views as against those of Needham. It answered all the latter's fears about the possible alteration of the air in Spallanzani's experiments; and it finally destroyed Gay-Lussac's hypothesis on the role of oxygen in the procedure for making Appert's preserves and in alcoholic fermentation.

However, on the latter point there were still some doubts. For in this same work by Dr. Schwann, besides the experiment with the meat juice (an experiment concerned with the cause of putrefaction), there was another (relating to alcoholic fermentation) which must be recalled. The author filled four flasks with a solution of cane sugar mixed with brewers' yeast. Then, after having stoppered them securely, he placed them in boiling water and later inverted them in a pneumatic trough filled with mercury [so that when the stoppers were subsequently removed the mercury cut off communication between the flasks and the atmosphere]. After the flasks had cooled he let ordinary air pass into two of them, and to the other two he admitted calcined air [air that had previously been passed through a red hot tube]. At the end of a month fermentation was evident in the flasks that had received ordinary air, but neither then nor after two months was fermentation detectable in the other two flasks. However on repeating these experiments I found, says he, that they do not always go so well. Sometimes fermentation does not appear in any of the flasks (for instance, when they have been kept too long in the boiling water); and sometimes, on the other hand, the liquid ferments even in the flasks that have received calcined air.

In summary, Dr. Schwann's experiment on the putrefaction of the broth [meat juice] is very clear. But concerning the alcoholic fermentation (the only fermentation about which much was known in 1837, when Schwann did his work) the learned physiologist's experiments gave contradictory results. In the meantime it had just been learned, from the observations of Cagniard de Latour [Charles Cagniard de la Tour

(1777–1859)] and from those of Schwann himself, that vinous fermentation is caused by an organized [living] ferment.

The obscurities of the situation, in so far as alcoholic fermentation was concerned, were greatly increased when, subsequently, chemists studied a great many fermentations in which no organized ferment could be discovered. The cause of these fermentations was universally attributed to the action of contact, to phenomena of impulsion or communicated movement produced by dead nitrogenous matter in process of decay. Be this as it may, here is the conclusion that Dr. Schwann drew from the experiments that I have just described. "It is not oxygen, or at least not just the oxygen of atmospheric air, which occasions alcoholic fermentation and putrefaction," says he, "but a principle present in ordinary air, a principle destroyed by heat."

The caution with which this conclusion is phrased should be noted. One easily sees, from certain passages in his work, that Dr. Schwann was inclined to believe that by heat he had destroyed germs; but his definitive conclusion could not and does not go as far as that assertion. In reporting his experiments the adversaries of the doctrine of spontaneous generation have often affirmed that the use of heat has no other purpose than the killing of germs; but this was only a hypothesis. As Dr. Schwann well said, these experiments prove only that it is not oxygen, or at least not oxygen alone, which is the cause of putrefaction and vinous fermentation, but an unknown *something* that heat destroys. And especially in the case of vinous fermentation there was but slight evidence that one had to postulate any other cause than that indicated by Gay-Lussac: namely, atmospheric oxygen itself.

Dr. Schwann's experiments have been repeated and modified by a number of investigators. Ure and Helmholtz [Andrew Ure (1778–1857), a Scotch physician and chemist; Hermann L. F. von Helmholtz (1821–1894), then at the threshold of a distinguished career as physicist, physiologist, and philosopher] have confirmed his results by experiments analogous to his. Instead of calcining the air before bringing it in contact with Appert's preserves, Schulze [Franz F. Schulze (1815–1873), professor of chemistry at the University of Rostock] passed the air through chemical reagents: concentrated potassium hydroxide and sulfuric acid. Schroeder and von Dusch [Theodor von Dusch; and Heinrich G. F. Schröder (1810–1885), schoolmaster and chemist] conceived the idea of filtering the air through cotton — instead of modifying it by high temperature, as in the procedure of Dr. Schwann, or by active chemical reagents, as in Schulze's procedure. Schroeder and von Dusch's first memoir appeared in 1854, the second in 1859. These are excellent works; and they have, furthermore, the historical merit of showing the state in 1859 of the question that now concerns us.

It had been known for a long time, ever since the first discussions of spontaneous generation, that a fine gauze (like that used with such success by Redi in his researches on the origin of larvae in putrefying meat) was sufficient to prevent, or at least to modify strikingly, the putrefaction of infusions. This very fact was among those emphasized by the adversaries of the doctrine of spontaneous generation.* Schroeder and von Dusch were, no doubt, guided by these facts; and above all (as they expressly state) by the ingenious experiments of Loewel [the dye chemist Henri Loewel, (1795–1856)], who recognized that, after it has been filtered through cotton, ordinary air is incapable of initiating the crystallization of sodium sulfate.†

Schroeder and von Dusch proceeded as follows. The organic matter was placed in a round-bottomed glass flask. Two tubes, bent at right angles, passed through the stopper of the flask. One of these tubes was connected with a water aspirator [a suction pump powered by water], the other with a large tube, one inch in diameter and twenty inches long, filled with cotton. When the connections had been made, the cock on the aspirator closed, and the organic matter placed in the flask, the latter was heated to a boil. Boiling was maintained for a time long enough for all the connecting tubes to be strongly heated by the steam. Then the cock on the aspirator was opened and the aspirator was kept going day and night [so that a stream of filtered air was maintained in the apparatus].

The results of the first tests conducted in this manner were as follows. Schroeder and von Dusch worked with: (1) meat with added water, (2) must of beer, (3) milk, (4) meat without water. In the first two cases air filtered through cotton left the liquids intact, even after several weeks. But the milk curdled and spoiled as promptly as in ordinary air, and the meat without water promptly began to putrefy. "From these experiments it seems clear," say Schroeder and von Dusch, "that there are some spontaneous decompositions of organic substances which need for their initiation only the presence of gaseous oxygen: for instance, the putrefaction of meat without water, the putrefaction of the casein of milk, and the transformation of milk sugar into lactic acid (lactic fermentation). But besides these there are other phenomena of putrefaction and fermentation that are incorrectly placed in the same category as the preceding. Such phenomena as the putrefaction of meat juice and alcoholic fermentation need for their initiation something besides oxygen. They need those unknown materials, present in atmospheric air, which are destroyed by heat according to Schwann's experiments and, according to ours, by the filtration of atmospheric air through cotton . . . Since so many questions still remain to be decided by experimentation, we shall abstain from drawing any theoretical conclusion from our experiments."

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* [At this point Pasteur cites, in a footnote, "an excerpt from the work of Baker, a member of the Royal Society of London, entitled: *The Microscope Made Easy* (London, 1743)."]

† [When solutions of most substances are made sufficiently concentrated, by evaporation, the behavior normally observed is a progressive deposition of the dissolved materials. But with solutions of certain substances, of which sodium sulfate is one, a special phenomenon of "supersaturation" is observed. From a solution of such a substance no crystalline deposit appears even when the solution contains far more of the substance than it can normally dissolve. However, crystallization *can* be initiated by the addition to the solution of a minute crystal of the substance concerned. Around this "seed" there is a sudden and voluminous deposition of the crystalline substance previously dissolved in the supersaturated solution. Solutions of many substances can be "seeded," and so made to crystallize, by minute dust particles (floating in the air) which we suppose have shapes or structures analogous to those of the substances concerned. Thus, for example, recent attempts at artificial rain-making have sought to initiate the production of fine ice crystals (thought to be the precursors of rain), by "seeding" air supersaturated with water vapor with dusts of substances (for example, silver iodide) which have crystal structures analogous to that of ice. But these attempts have not been uniformly successful, our knowledge of the phenomena involved in the relief of supersaturation is still imperfect, and Loewel's study is but one early chapter of a story not yet completed.]

Schroeder returns alone to this subject, in 1859, in a memoir that also considers the cause of crystallization. This new work did not lead its author to any completely definite conclusions. He reports some new organic liquids that do not putrefy when placed in contact with filtered air. Such liquids are urine, starch paste, and the various constituents of milk taken separately. But he adds egg yolk to the list of substances that, like milk and meat without water, putrefy in air filtered through cotton.

"I shall not hazard an attempt at a theoretical explanation of these facts," says Schroeder. "It might be supposed that fresh air contains an active substance — a substance destroyed by heat or held back by cotton — which initiates the phenomena of alcoholic fermentation and putrefaction." Then he adds: "Should this active substance be regarded as formed of microscopic organisms [germs] disseminated in the air? Or is it indeed a chemical substance still unknown? I do not know."

Later he comes to the phenomena of crystallization in open air, in heated air, and in air filtered through cotton. According to him, these phenomena seemed so analogous to the phenomena of putrefaction that he could not refrain from attributing them to a common cause until then entirely unknown.

"As far as crystallization is concerned," he continues, "the *inductive* action of the air [the action by which air initiates crystallization] does not seem to be entirely stopped by the cotton, but only weakened. The filtration of the air through cotton prevents the crystallization only of certain supersaturated solutions, but there are other solutions that are always affected by the filtered air." Then he remarks that the results of his studies of putrefaction and fermentation are parallel to those connected with crystallization. For there are substances which remain unaltered in the filtered air while others, such as milk, decompose in its presence. Thus air filtered through cotton only partially loses its *inductive* force for putrefaction and fermentation.

I have intentionally reviewed in detail these very careful researches, since they give an accurate picture of the difficulties that existed in 1859. These difficulties besieged all impartial minds, free from preconceived ideas, that sought to form a well-founded opinion on this serious question of spontaneous generation. It may be said that at this time all those who believed that the question had been resolved were ill-informed of its history.

Spallanzani had not triumphed over the objections of Needham; and Schwann, Schulze, and Schroeder had only demonstrated that in atmospheric air there exists an unknown principle essential for life in infusions. Those who declared that this principle was nothing but germs had no more proofs for their opinion than those who believed that it might be a gas, a fluid, noxious effluvia, etc., and who consequently were inclined to believe in spontaneous generation. The conclusions of Schwann and Schroeder are in this respect [that is, with regard to their inconclusiveness] perfectly clear. The very terms of these conclusions provoked doubt, and so served the proponents of the doctrine of spontaneous generation. Also, the experiments of Schwann, Schulze, and Schroeder succeeded only with certain liquids. Moreover, as I shall soon report, they failed almost uniformly, and with all liquids, when they were conducted with a pneumatic trough filled with mercury. Nobody understood the reason for this failure, and nobody could discover any cause of error.

Thus nobody could point out the true cause of error in Pouchet's experiments when, after the investigations I have just discussed, that skillful naturalist of Rouen (a corresponding member of the Academy of Sciences) announced to the Academy the results on which he believed he could establish the principles of heterogeneity [spontaneous generation] in a definitive manner. And soon, realizing how much remained to be done, the Academy offered a prize for a dissertation on the following subject: *Attempts by well-conceived experiments to throw new light on the question of spontaneous generation.* [The prize was won by Pasteur with the "Memoir" you are now reading.]

The problem then seemed so obscure that Biot, whose kindness with regard to my work has always been unfailing, expressed his regret at seeing me engaged in these researches. Claiming my deference to his advice, he exacted from me a promise to abandon the subject if, at the end of a specified time, I had not mastered the difficulties that were then perplexing me. At about the same time Dumas, who has often joined with Biot in showing kindness to me, said: "I would not advise anyone to spend too long on this subject." [Courage was required to resist the advice of Jean Baptiste Biot (1774–1862), mathematician, physicist, astronomer, and then an elder statesman of French science; and of Jean Baptiste Dumas (1800–1884), Pasteur's respected teacher and the most influential French chemist of that day.]

What need had I to concern myself with this subject? Twenty years ago chemists came upon a collection of truly extraordinary phenomena, designated by the generic name *fermentations.* All require the simultaneous presence of two substances: one, such as sugar, called *fermentable*; the other, always an albumen-like material, called *nitrogenous.* Here is the theory of fermentation that was universally accepted: on exposure to air the albuminous materials undergo a change, a special oxidation of unknown nature, which gives them the character of a *ferment*—that is to say, the property of subsequently acting, through contact, on fermentable substances.

There was certainly one ferment, the oldest and most remarkable of all, that was known to be a living organism: brewers' yeast. But, even after careful examination, no one had been able to recognize the existence of living organisms in any of the fermentations discovered more recently than the recognition (in 1836) of the fact that brewers' yeast is a living organism. Thus physiologists gradually abandoned, some with regret, Cagniard de Latour's hypothesis of a probable relation between the life of the ferment and its action as a ferment. The general theory [the contact theory of fermentation] was applied to brewers' yeast in such terms as these: "Brewers' yeast is active, not because it is a [living] organism, but because it has been in contact with air. It is the dead portion of the yeast, that which was alive and is in process of decay, which acts upon the sugar."

My studies led me to entirely different conclusions. That all fermentations properly so called—lactic, butyric, the fermentations of tartaric acid, of malic acid, of urea [in urine], the fermentation of sugar to mannite—were always associated with the presence and multiplication of living organisms. And instead of being an obstacle to the theory of fermentation, the life of yeast was, on the contrary, the very fact that made yeast fit into the usual role, and a prototype of all true ferments. According to my views albuminous materials are never ferments, but the food of ferments. The true ferments are living organisms.

That ferments arise from the contact of albuminous substances with oxygen gas is well known. Then, I said to myself, one of two things must be true. The true ferments being living organisms, if they are produced by the contact of albuminous materials with oxygen alone, considered merely as oxygen, then they are spontaneously generated. But if these living ferments are not of spontaneous origin, then it is not just the oxygen as such that intervenes in their production—the gas acts as a stimulant to a germ carried with it or already existing in the nitrogenous or fermentable materials. At this point, to which my study of fermentation brought me, I was thus obliged to form an opinion on the question of spontaneous generation. I thought I might find here a powerful support for my ideas on those fermentations which are properly called fermentations.

The researches that I am about to describe were, consequently, only a digression forced upon me by my studies of fermentation. It was thus that I was led to occupy myself with a subject that till then had taxed the skill and wisdom of naturalists only.

2. TRANSLATION OF EXCERPTS FROM REDI'S "EXPERIMENTS ON THE GENERATION OF INSECTS." *

It being thus, as I have said, the dictum of ancients and moderns, and the popular belief, that the putrescence of a dead body, or the filth of any sort of decayed matter engenders worms; and being desirous of tracing the truth in the case, I made the following experiment:

At the beginning of June I ordered to be killed three snakes, the kind called eels of Aesculapius. As soon as they were dead, I placed them in an open box to decay. Not long afterwards I saw that they were covered with worms of a conical shape and apparently without legs. These worms were intent on devouring the meat, increasing meanwhile in size, and from day to day I observed that they likewise increased in number; but, although of the same shape, they differed in size, having been born on different days. But all, little and big, after having consumed the meat, leaving only the bones intact, escaped from a small aperture in the closed box, and I was unable to discover their hiding place. Being curious, therefore, to know their fate, I again prepared three of the same snakes, which in three days were covered with small worms. These increased daily in number and size, remaining alike in form, though not in color. Of these, the largest were white outside, and the smallest ones, pink. When the meat was all consumed, the worms eagerly sought an exit, but I had closed every aperture. On the nineteenth day of the same month some of the worms ceased all movements, as if they were asleep, and appeared to shrink and gradually to assume a shape like an egg. On the twentieth day all the worms had assumed the egg shape, and had taken on a golden white color, turning to red, which in some darkened, becoming almost black. At this point the red, as well as the black ones, changed from soft to hard, resembling somewhat those chrysalides formed by caterpillars, silkworms, and similar insects. My curiosity being thus aroused, I noticed that there was some difference in shape between the red and the black eggs [pupae],[1] though it was clear that all were formed alike of many rings joined together; nevertheless, these rings were more sharply outlined, and more apparent in the black than in the red, which last were almost smooth and without a slight depression at one end, like that in a lemon picked from its stalk, which further distinguished the black egglike balls. I placed these balls separately in glass vessels, well covered with paper, and at the end of eight days, every shell of the red balls was broken, and from each came forth a fly of gray color, torpid and dull, misshapen as if half finished, with closed wings; but after a few minutes they commenced to unfold and to expand in exact proportion to the tiny body, which also in the meantime had acquired symmetry in all its parts.

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* [Translated from the Italian edition of 1688 by Mat Bigelow, Chicago, Open Court, 1909.]

[1] Throughout this work Redi uses the word "uova" where the context shows that pupa is meant. In this he followed Harvey, who called any embryonic mass an "egg."

Then the whole creature, as if made anew, having lost its gray color, took on a most brilliant and vivid green; and the whole body had expanded and grown so that it seemed incredible that it could ever have been contained in the small shell. Though the red eggs [pupae] brought forth green flies at the end of eight days, the black ones labored fourteen days to produce certain large black flies striped with white, having a hairy abdomen, of the kind that we see daily buzzing about butchers' stalls. These at birth were misshapen and inactive, with closed wings, like the green ones mentioned above. Not all the black eggs [pupae] hatched after fourteen days; on the contrary, a large part of them delayed until the twenty-first day, at which time there came out some curious flies, quite distinct from the other two broods in size and form, and never before described, to my knowledge, by any historian, for they are much smaller than the ordinary house-flies. They have two silvery wings, not longer than the body, which is entirely black. The lower abdomen is shiny, with an occasional hair, as shown by the microscope, and resembles in shape that of the winged ants. The two long horns, or antennae (a term used by writers of natural history), protrude from the head; the first four legs do not differ from those of the ordinary fly, but the two posterior ones are much larger and longer than would appear to be suitable for such a small body; and they are scaly, like the legs of the locusta marina; they are of the same color, but brighter, so red, in fact, that they would put cinnabar [the brilliant sulfide ore of mercury] to shame; being all covered with white spots, they resemble fine enamel work . . .

I continued similar experiments with the raw and cooked flesh of the ox, the deer, the buffalo, the lion, the tiger, the dog, the lamb, the kid, the rabbit; and sometimes with the flesh of ducks, geese, hens, swallows, etc., and finally I experimented with different kinds of fish, such as sword-fish, tuna, eel, sole, etc. In every case, one or other of the above-mentioned kinds of flies were hatched, and sometimes all were found in a single animal. Besides these, there were to be seen many broods of small black flies, some of which were so minute as to be scarcely visible, and almost always I saw that the decaying flesh and the fissures in the boxes where it lay were covered not alone with worms, but with the eggs from which, as I have said, the worms were hatched. These eggs made me think of those deposits dropped by flies on meats, that eventually become worms, a fact noted by the compilers of the dictionary of our Academy, and also well known to hunters and to butchers, who protect their meats in summer from filth by covering them with white cloths. [This is a good illustration of the significance of a practice of an artisan as a starting point for a scientific investigation.] Hence great Homer, in the nineteenth book of the *Iliad*, has good reason to say that Achilles feared lest the flies would breed worms in the wounds of dead Patroclus, whilst he was preparing to take vengeance on Hector.

Having considered these things, I began to believe that all worms found in meat were derived directly from the droppings of flies, and not from the putrefaction of the meat, and I was still more confirmed in this belief by having observed that, before the meat grew wormy, flies had hovered over it, of the same kind as those that later bred in it. [Here we see Redi formulating a working hypothesis.] Belief would be vain without the confirmation of experiment, hence in the middle of July I put a snake, some fish, some eels of the Arno, and a slice of milk-fed veal in four large, wide-mouthed flasks; having well closed and sealed them, I then filled the same number of flasks in the same way, only leaving these open. [The control experiment!] It was not long before the meat and the fish, in these second vessels, became wormy and flies were seen entering and leaving at will; but in the closed flasks I did not see a worm, though many days had passed since the dead flesh had been put in them. Outside on the paper cover there was now and then a deposit, or a maggot that eagerly sought some crevice by which to enter and obtain nourishment. Meanwhile the different things placed in the flasks had become putrid and stinking . . .

Not content with these experiments, I tried many others at different seasons, using different vessels. In order to leave nothing undone, I even had pieces of meat put under ground, but though remaining buried for weeks, they never bred worms, as was always the case when flies had been allowed to light on the meat. One day a large number of worms, which had bred in some buffalo meat, were killed by my order; having placed part in a closed dish, and part in an open one, nothing appeared in the first dish, but in the second worms had hatched, which changing as usual into eggshape balls [pupae], finally became flies of the common kind . . .

. . . Although I thought I had proved that the flesh of dead animals could not engender worms unless the semina of live ones were deposited therein, still, to remove all doubt, as the trial had been made with closed vessels into which the air could not penetrate or circulate, I wished to attempt a new experiment by putting meat and fish in a large vase closed only with a fine Naples veil, that allowed the air to enter. For further protection against flies, I placed the vessel in a frame covered with the same net. I never saw any worms in the meat, though many were to be seen moving about on the net-covered frame. These, attracted by the odor of meat, succeeded at last in penetrating the fine meshes and would have entered the vase had I not speedily removed them. It was interesting, in the meantime, to notice the number of flies buzzing about which, every now and then, would light on the outside net and deposit worms there. I noted that some left six or seven at a time there, and others dropped them in the air before reaching the net . . .

3. CERTAIN OF PASTEUR'S EXPERIMENTAL RESULTS AS REPORTED IN CHAPTER IV OF HIS "MEMOIR" OF 1862

In the second chapter of Pasteur's "Memoir," he reports experiments in which a stream of ordinary air is drawn through a wad of guncotton (nitrated cellulose). The wad is then dissolved in a mixture of alcohol and ether and the particles that remain are examined. Under a microscope some of the particles had "clearly defined outlines much like the spores of common molds." Others resembled certain of the microörganisms (*Infusoria*) commonly found in fermenting or putrefying liquids. Of course, any living organisms that were thus caught in the guncotton were killed by the action of the alcohol-ether mixture. Therefore, this evidence for the presence of "germs" in city air was, to say the least, indirect.

In the third chapter Pasteur presents better evidence for the existence of "fertile germs" along with dust in city air. Here he borrows a technique from Dr. Schwann's earlier work and "sterilizes" his air (the term was invented later) by passing it through a red-hot tube. This "calcined air," Pasteur found, could be introduced into a flask containing "sugared yeast water" without inducing any growths in the medium, while ordinary city air, when similarly introduced, almost invariably produced growths. Pasteur's medium, it must be noted, was a 10-percent solution of sugar in yeast water, that is, water which had extracted some of the protein and mineral matter from brewers' yeast. Prepared by treating yeast with boiling water, Pasteur's yeast water contained, of course, no living yeast cells. The sugared yeast water was placed in a round-bottomed flask of 250- to 300-cubic-centimeter capacity, with a long neck connected to a supply of calcined air. The liquid was boiled for two or three minutes, and the neck was then sealed off. The contents of such flasks were sterile, as shown by the complete absence of growths, even after many days in an incubator at 30°C (86°F).

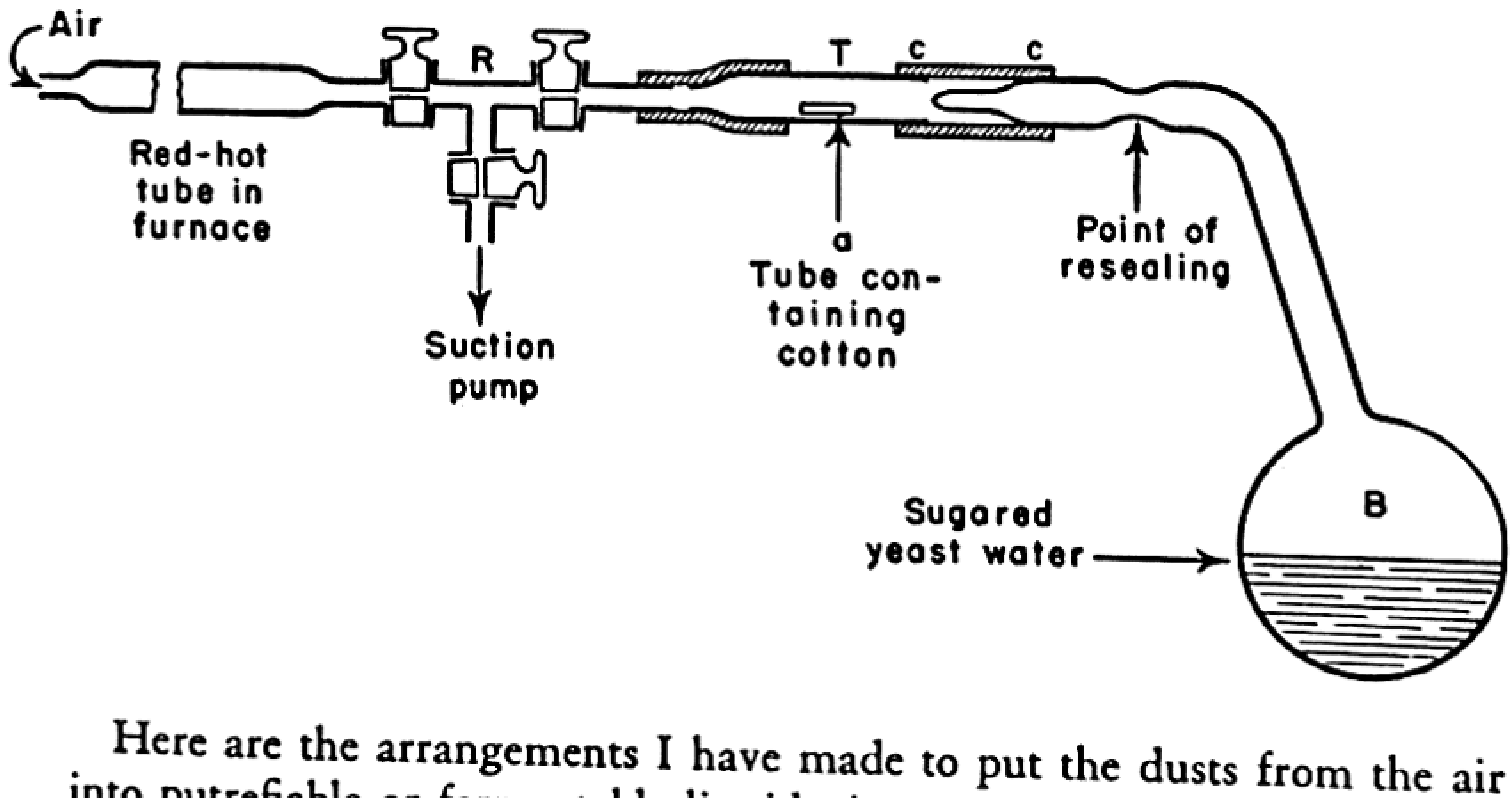
"I declare with complete sincerity," wrote Pasteur, "that I have never had a doubtful result from an experiment of this sort. Sugared yeast water boiled for two or three minutes and then exposed to calcined air does not change [ferment] even after standing for 18 months at a temperature of 25° to 30°. While if ordinary air is admitted then, after a day or two of incubation, alterations begin to appear and the liquid becomes full of bacteria or covered with molds."

With this background of information about Pasteur's use of the "calcined-air" technique, we are in a position to read one of his most significant reports. Chapter IV of his "Memoir" of 1862 deals with the sowing of dusts collected from the atmosphere into sterile sugared yeast water.

SOWING OF THE DUSTS, WHICH EXIST SUSPENDED IN THE AIR, INTO LIQUIDS WHICH ARE SUITABLE FOR THE DEVELOPMENT OF THE LOWER ORGANISMS.

The results of the experiments of the two preceding chapters have shown us: (1) that there are always in suspension in ordinary air organized particles completely like the germs of the lower organisms; (2) that sugared yeast water, a liquid easily alterable in ordinary air, remains intact, limpid, never giving birth to infusoria or molds when it is left in contact with air that has been previously heated.

Given these facts, let us try to discover what will happen if we introduce, into the water containing sugar and albumin, the dusts that we have already learned to collect [by drawing a stream of ordinary air through a glass tube plugged with a porous material, like cotton]—taking care that nothing else is introduced and that the liquid is in contact with this same heated air . . .



Here are the arrangements I have made to put the dusts from the air into putrefiable or fermentable liquids, in an atmosphere of air that has been heated.

Let us take our large flask containing sugared yeast water and calcined air. I shall suppose that the flask has been kept in a chamber at 25° or 30° for one or two months without having shown any perceptible alteration—a manifest proof of the inactivity of the heated air with which the flask has been filled under ordinary atmospheric pressure.

The constricted neck of the flask remaining sealed, I connect it by means of a rubber tube to an apparatus arranged as shown in the accompanying figure. *T* is a hard-glass tube with internal diameter of 10 to 12 millimeters. In this I have placed a little tube (*a*) of small diameter and free to slip in the large tube. The little tube has open ends and contains part of one of the small wads of cotton loaded with dust. *R* is a brass tube in the form of a T and provided with stopcocks. One of these stopcocks is connected with the air pump [suction pump], the second with a platinum tube heated red hot, the third with tube *T*. The rubber tube connecting tube *T* with flask *B* is shown at *cc*.

When all the parts of the apparatus have been assembled and the platinum tube has been brought to red heat by a gas furnace, the vacuum pump is started after the stopcock leading to the platinum tube has been closed. This stopcock is later opened in such a way that the calcined air enters the apparatus very gradually. The evacuation and the admission of the calcined air are alternately repeated ten to twelve times. The little tube carrying the cotton is thus filled with heated air, even in the smallest interstices of the cotton, but the cotton retains its dust. This done, I break the sealed tip of flask *B* from outside the rubber tube *cc*, without loosening the latter's bindings [to flask *B* and tube *T*]. Then [by lowering flask *B*] I make the little tube containing the dust slip into the flask. Finally I use a [glassblowers'] lamp to reseal the drawn-out neck of the flask, which is then once again returned to the incubator. Now it happens without fail that growths begin to appear in the flask after 24, 36, or 48 hours at the most.

This is precisely the time required for the appearance of the same growths in sugared yeast water when it has been exposed to the contact of ordinary air. Here are the details of several experiments.

In the first days of November, 1859, I prepared several flasks of 250-cubic-centimeters capacity, holding 100 cubic centimeters of sugared yeast water and 150 cubic centimeters of heated air. The flasks remained in an incubator, at a temperature around 30°, until January 8, 1860. On that day, at about 9 A.M., I used the apparatus shown in the figure to introduce into one of these flasks a portion of a wad of cotton charged with dust, which had been collected as explained in Chapter II.

On January 9th, at 9 A.M., the liquid in the flask showed no change. At 6 P.M. on the same day little tufts of mold could be seen very distinctly, growing from the tube containing the dust. The liquid remained perfectly limpid.

On January 10th, at 5 P.M., the liquid still maintained its perfect limpidity, aside from the silky tufts of mold. On the sides of the flask I now perceived a great number of white streaks which, when the flask was held to the light, displayed several iridescent colors.

On January 11th the liquid had lost its limpidity. It was entirely turbid, so much so that the tufts of the Mycelium could no longer be distinguished.

Then, using a file, I opened the flask and studied with a microscope the different growths that had originated therein.

The turbidity of the liquid was due to a crowd of little bacteria, of the smallest dimension, very rapid in their movements, pirouetting in a lively manner or balancing themselves. The silky tufts were formed by a Mycelium with branched tubes.

Finally, the sort of pulverizable precipitate, that appeared on January

10th in the form of white streaks, is made up of very fine "Torulacée." This "Torulacée" is frequently found in sugared albuminous liquids. It develops, for example, in beet juice which has been made a little acid, and in the urine of diabetic persons. It could easily be confused with brewers' yeast, which it very much resembles in its mode of development. However, the diameter of its globules is appreciably less than the diameter of the yeast globules — smaller by one third or even by one half. The globules of this "Torulacée" are only slightly granular, and are more translucid than the globules of brewers' yeast. The nucleus, when it is visible, is single and very clear. The globules of this "Torulacée" multiply by budding and resemble the branched form of brewers' yeast in process of multiplication.

Thus, here are three growths produced under the influence of the dusts which were sowed — growths of the same order as those that originate in these same sugared albuminous liquids when the liquids are left in contact with ordinary air.

On January 17th I introduced dust into two more of the flasks containing sugared yeast water which had remained unchanged since the month of November.

On the morning of the 19th the liquid in one of the flasks was very turbid. However, there was no appearance of Mycelium. The liquid in the other flask was still very limpid. There was no appearance of organized growth.

At 5 P.M. on the same day the first flask was in the same state; the turbidity had simply increased. As to the other, the limpidity of its liquid remained perfect, but a tuft of Mycelium was growing from the small tube containing the dust, and entirely wreathed its end.

On the 20th the state of the first flask had not changed perceptibly. The mold in the second had developed greatly, and a new mold had formed within the liquid. Besides, the limpidity of the liquid seemed slightly changed.

On the 21st the liquid in the second flask was almost as turbid as that in the first, and the tufts of Mycelium had not increased since the day before — that is to say, not since the turbidity had appeared in the bulk of the liquid.

On the 22nd and 23rd of January the tufts of Mycelium remained the same. We shall see that the stoppage of the development of the Mycelium must be attributed to the presence of the Infusoria that rendered the liquid turbid. These Infusoria seize upon the dissolved oxygen, and so deprive the Mycelium of one of its most essential nutrients. This result is constant and explains why in the first flask, where Infusoria were the first growth to develop, no other organized growths appeared . . .

One might well ask whether the cotton used in these experiments has not had some influence on the results, since the cotton is itself an organic material [that is, a material that is uniquely the product of a living organism]. It is most important to know what would happen if the same manipulations were performed with flasks prepared as before, but without adding the dusts from the air. In other words, did the manipulations required to introduce the dusts have any influence? This we must know.

In order to answer these questions, I replaced the cotton by asbestos. Wads of asbestos, through which a current of ordinary air had been passed for several hours, were introduced into the flasks, following the same procedure as above, and they gave results of exactly the same sort as those just reported. But with wads of asbestos previously calcined and not charged with dust, *or charged with dust and afterwards heated, no turbidity, no Infusoria, no plants of any sort are produced. The liquids remain perfectly limpid.* [The phrases describing what appears to be the most convincing evidence have been italicized by the translator.] I have repeated these comparative experiments a great many times, and I have always been surprised by their clearness and perfect reproducibility. It would seem, indeed, that experiments of this delicacy should sometimes yield contradictory results, due to accidental sources of error. But, just as the sowing of dusts has always furnished living organisms, never once did my blank experiments show such growths.

With such results, confirmed and extended by those of the following chapters, I consider this as mathematically demonstrated: that all the organisms which appear in sugared albuminous solutions that have been boiled and then exposed to ordinary air have their origin in the solid particles suspended in the atmosphere.

But, on the other hand, we have seen in Chapter II that these solid particles include — amid a multitude of amorphous fragments of calcium carbonate, silica, soot, bits of wool, etc. — organized corpuscles that are indistinguishable from the little spores of the growths we have seen formed in sugared albuminous liquids. These corpuscles are therefore the fertile germs of these growths.

We may conclude, moreover, that if an Appert preserve formed of a sugared albuminous liquid, such as the must of raisins, does not change when it is brought in contact with air that has been heated (as Dr. Schwann was the first to observe) it is because the heat has destroyed the germs which the air was carrying. All the adversaries of spontaneous generation foresaw this. All I have done is to supply sure and decisive proofs. Nonprejudiced minds are now obliged to reject completely any idea that there exists in air a more or less mysterious principle, gas, fluid, ozone, etc. having the property of arousing life in infusions.

One might consider at this point a very interesting question, to which I shall return in a special publication, which will not fail to surprise the reader. Nothing is better adapted for the production of alcoholic fermentation than the liquid studied in the preceding pages. Sugared yeast water — like grape must, beer must, beet juice, etc. — is a liquid which readily ferments when exposed to ordinary air. But in a considerable number of experiments carried out as previously described, no fermentation of the sugared liquid could be obtained by sowing in it dusts from

> the air. (I shall show later that this peculiarity arises from the relation between the volumes of air and liquid present in my experiments.)
>
> I should here emphasize that there is nothing more contrary to the truth than the assertions often repeated by the partisans of the doctrine of spontaneous generation: "That the appearance of the first organisms is always preceded by phenomena of fermentation or putrefaction . . . that the formation of the animalcules in the macerated material is a consequence of the release of the gases formed by the decomposition of the substances used, and that it is only after these phenomena that a special film (pellicle) forms at the surface of the liquid" (Pouchet, 1859). Also, when some one speaks to me of the motion of fermentation which I set up in my liquids by sowing dust in them — *the fermentative movement necessary for the development of the germinal forces* [forces génésique] — I recognize only vague words to which experience teaches me to lend no real sense.

The observations referred to in the last paragraphs of this section of Pasteur's "Memoir" have little to do with the question of spontaneous generation. They are preliminary to those studies which led Pasteur to state his famous doctrine that "fermentation is life without oxygen." With some modifications this statement is true today. Many microorganisms, which in the presence of oxygen derive the energy for their growth by oxidizing the substrate (that is, sugar) to carbon dioxide and water, may in the absence of oxygen obtain energy from fermentative processes. Thus, the change of sugar to alcohol and carbon dioxide yields the energy for the growth of yeast cells under anabolic (that is, oxygen-free) conditions.

As early as 1862, then, Pasteur saw the importance of the presence or absence of oxygen for fermentation. It is somewhat ironic that he failed to guess that for some microörganisms oxygen is essential for any growth and, further, that some of these aerobic bacteria form highly heat-resistant spores. Ignorant of these facts, Pasteur presents in Chapter VII of his historic "Memoir" evidence that he regards as proof that, by itself, a small quantity of air will not produce growths in a sterile medium. With sugared yeast water his experiments worked; with Pouchet's hay infusion they did not, for reasons already explained to the reader in the General Introduction.

4. TRANSLATION OF A FURTHER PORTION OF PASTEUR'S "MEMOIR" OF 1862

CHAPTER VII

IT IS NOT TRUE THAT THE SMALLEST QUANTITY OF ORDINARY AIR IS SUFFICIENT TO PRODUCE IN AN INFUSION ORGANIZED LIFE CHARACTERISTIC OF THAT INFUSION — EXPERIMENTS WITH AIR FROM DIFFERENT LOCALITIES . . .

In the historical part of this Memoir I have already referred to the influence, on the subject with which we are concerned, of Gay-Lussac's celebrated investigation of the air in Appert's preserves, and of the illustrious natural philosopher's interpretation of his experiments. Here are his own words:

"By analyzing the air in bottles in which substances have been well preserved, one can convince oneself that it contains no oxygen and, consequently, that the absence of this gas is a necessary condition for the preservation of animal and vegetable substances."

There can be no doubt that the air from the preserves studied by Gay-Lussac was free of oxygen. No one would dare to suspect the accuracy of an analysis of air made by Gay-Lussac [who was the preeminent gas analyst of his time]. However, today it cannot be doubted — even though no one has (as far as I know) repeated exactly these experiments of Gay-Lussac's — that Appert's preserves can contain oxygen, especially when they are newly prepared. From the analyses of air that I have reported elsewhere in this Memoir, it follows that the oxygen of air made inactive by heat, by Schwann's method, combines directly with organic materials, producing carbonic acid [carbon dioxide]. This is a very slow action. Nevertheless, direct oxidation exists: it cannot be denied. This oxidation may be more evident in Appert's preserves when they are being prepared, because of the elevated temperature. In all cases, if the preparation leaves any oxygen in the preserves, this gas will disappear little by little because of the direct oxidation of which I have just spoken. There is a circumstance which must do much to minimize or to reduce to zero the quantity of oxygen remaining in Appert's preserves: this circumstance is the ratio of the volume of air to the volume of organic matter. The preserves always contain very little air and a great deal of organic matter — a circumstance very favorable to the completion of the oxidative process [that is, to the complete exhaustion of the small amount of oxygen available in the container]. But, I repeat, nothing would be easier than the preparation of preserves in which oxygen remained, and there is room to believe that they often contain oxygen. Schwann's experiment leaves no doubt on this point.

This is why Gay-Lussac's interpretation of his analyses — namely, *that the absence of oxygen is requisite for preservation* [of organic materials] — is totally erroneous. However, not everyone has been able to draw the line between the truth of Gay-Lussac's observations and the error of his interpretation of them. Dr. Schwann should justly be regarded as the author of the true theory of Appert's procedures. Appert's preserves remain unspoiled in the presence of heated air: that is his discovery. The secret of their preservation is thus to be found not in the absence of oxygen,[1] but in the destruction by heat of a principle contained in ordinary air.

But there is one implication of Gay-Lussac's experiments which Schwann's discovery did not discredit, an implication that Schwann's discovery rather tended to confirm. This implication has not been contested by the adversaries of the doctrine of spontaneous generation, and on it the partisans of this doctrine justly rest one of their principal objections. This implication is that the smallest quantity of common air, brought in contact with an infusion, soon produces therein the growth of the Mucedines and Infusoria that commonly occur in that infusion. [This *is* true if the spores of aerobic bacteria are present!]

This point of view has always been supported (at least indirectly) by the set habit considered indispensable by investigators — the use of infinite precautions in their experiments to exclude ordinary air. We have seen that sometimes they recommend that the common air be heated; sometimes they treat it with active chemicals; often they take the preliminary step of treating all the components with steam at 100° (Spallanzani's experiment); and finally, on other occasions, they work with an artificial atmosphere. And if, under one of these various conditions, it happens that the experiment results in the production of organized life, then they do not hesitate to affirm that the operator has not known how to avoid completely the hidden influence of a small portion of ordinary air — however small it may be.

From this the partisans of the doctrine of spontaneous generation hasten on to a quite reasonable deduction. If the smallest quantity of ordinary air suffices for the production of organisms in any medium whatsoever, and if these organisms do not originate spontaneously, then

[1] Although the absence of oxygen is not involved in the explanation of the Appert process, one should not conclude that in practice there would be no danger if much air were left in the preserves. For if the heat has not destroyed all the germs of Infusoria and Mucedines, carried by the air or the materials to be preserved, those germs that are still fertile could develop if oxygen were present. While in the absence of oxygen they will develop no more than if they had really been killed. But I believe that what is always most to be feared (and particularly when little oxygen is present) is the presence of the germs of animal or vegetable ferments that can live without air. Such germs must necessarily be killed by heat [for, if they are not, they will produce fermentation even in the absence of oxygen]. I am persuaded that this is the danger most to be feared by the manufacturer of preserves. And I am inclined to believe that, for example, the animalcules known as the butyric infusoria (which I have recently described) develop in certain preserves which have been badly prepared.

it follows necessarily that in this indefinitely small portion of common air there must be the germs of a multitude of different organisms. Finally, if things are thus, then ordinary air must, to use M. Pouchet's expressions, be clogged with organic matter, forming a dense fog.

This reasoning is surely very cogent. It would be even more so if it had been established that the lower forms of life are really as distinct as they appear to be, and consequently come from different germs. This is probable, but it has not been proved.

Here then is a serious and apparently well-founded difficulty. But does it not arise in exaggerations and in more or less erroneous observations? Is it true, as has been supposed, that there is in the earth's atmosphere a continuity of cause of the generations said to be spontaneous? *Is it certain that the smallest quantity of ordinary air is sufficient to produce organized life in all infusions?* [The translator has italicized Pasteur's statement of the central issue.]

The following experiments answer all these questions.

In a series of flasks of 250 cubic-centimeter capacity I place the same putrescible liquid (yeast water; the same with sugar; urine, etc.), the liquid occupying about a third of the total volume. I taper the necks of the flasks with a glassblowers' lamp, I boil the liquid, and I seal off the tapered ends while the boiling is going on. A vacuum is thus formed in the flasks [since the steam has driven out all the air]. Then I break off the sealed tips of the flasks in a specified location. Ordinary air rushes in violently, carrying with it all the dusts that it holds in suspension and all the known or unknown principles associated with it. Then, barely touching them with a flame, I immediately reseal the flasks, and I place them in an incubator at 25° or 30° — that is to say, at the most favorable temperature for the development of animalcules and Mucors.

Here are the results of these experiments, results inconsistent with the principles generally accepted and, on the contrary, in perfect agreement with the idea of a dissemination of germs.

Generally the liquid changes after a very few days, and a great variety of organisms can be seen growing in the flasks . . . But, on the other hand, it happens frequently (several times in each series of trials) that the liquid remains absolutely unchanged, however long it is left in the incubator, just as if calcined air had been admitted . . .

The remainder of this chapter of Pasteur's "Memoir" is taken up with a lengthy record of many experiments performed under a great variety of conditions. Pasteur had developed a method, it should be noted, for testing the assumption referred to in the opening paragraphs of this section of his paper. He raised the question: "Are we perfectly sure that the smallest quantity of ordinary air is sufficient to produce organized life in any infusion?" Then he proceeded to devise the apparatus necessary to answer this question and went on to carry out the tests. (One might say that Pasteur put forward the working hypothesis that the smallest quantity of ordinary air is sufficient to produce organized life in any infusion, and proceeded to test the deduction by specific experiments.) His results can be summed up in a few words. If a number of flasks are opened in any given locality, then sealed off and placed in an incubator, the subsequent appearances are not uniform. Almost without exception the contents of some flasks remain unchanged, while the contents of others soon show evidence of fermentation. Clearly the variable is the nature of the small amount of air that rushes in when the flasks are opened. This in turn depends on the locality in which they are opened. In short, Pasteur had demonstrated what he called the "discontinuity of the causes of so-called spontaneous generation." If all the flasks opened in all localities had shown evidence of fermentation, then one could have maintained that living organisms develop spontaneously when oxygen is admitted. This was, in fact, what his rival Pouchet claimed.

Both Pasteur and Pouchet assumed that boiling water would kill all living organisms (or their "seeds"). This is erroneous. Both assumed that "sugared yeast water" and hay infusion were equivalent in these experiments. This was unwarranted. Pasteur assumed that only the presence or absence of microörganisms in the entering air would affect the subsequent development of living organisms. Pouchet assumed that only the presence or absence of oxygen determined the subsequent events in the flasks in question. Actually *both* variables were involved in the case of hay infusion that had been heated only to boiling. The clarification of the matter, it should be noted, rested on subsequent experimental findings related to the controversy but not directly concerned with it. One had to go beyond the experiment that seemed designed to give a yes or no answer to the logical deductions from the grand working hypothesis. New modes of experimentation had to be developed before a baffling contradiction could be cleared up. Some idea of how this was done can be obtained from Tyndall's paper which now follows.

5. TYNDALL'S ARTICLE ON SPONTANEOUS GENERATION

The following is part of an article first published in 1878 in *The Nineteenth Century*. This article, subsequently reprinted in Tyndall's influential *Essays on the Floating Matter in the Air in Relation to Putrefaction and Infection* (London, 1881), presents a summary account of experiments on spontaneous generation originally reported by Tyndall in 1876 and 1877 in the *Philosophical Transactions of the Royal Society*. The material enclosed in brackets has been added by the editor of this case history.

> During the ten years extending from 1859 to 1869, researches on radiant heat in its relations to the gaseous form of matter occupied my continual attention. When air was experimented on, I had to cleanse it effectually of floating matter, and while doing so I was surprised to notice that, at the ordinary rate of transfer, such matter passed freely through alkalis, acids, alcohols, and ethers. The eye being kept sensitive by darkness, a concentrated beam of light was found to be a most searching test for suspended matter both in water and in air — a test indeed indefinitely more searching and severe than that furnished by the most powerful microscope. With the aid of such a beam I examined air filtered by cotton wool; air long kept free from agitation, so as to allow the floating matter to subside; calcined air, and air filtered by the deeper cells of the human lungs.

Tyndall had discovered that the last portion of a breath expelled from the lungs is remarkably free from suspended dust. Shortly afterward the full significance of this discovery was brought home to him by a statement made by Joseph Lister (1827–1912), who, by his introduction of the antiseptic method in surgery, made one of the earliest and most striking applications of the germ theory of disease which grew out of Pasteur's microbiological studies of fermentation and spontaneous generation.

Lister stated that: "I have explained to my own mind the remarkable fact that in simple fracture of the ribs, if the lung be punctured by a fragment, the blood effused into the pleural cavity, though freely mixed with air, undergoes no decomposition . . . (if the blood in the pleura were to putrefy, it would infallibly occasion dangerous suppurative pleurisy). Why air introduced into the pleural cavity through a wounded lung should have such wholly different effects from that entering directly, through a wound in the chest, was to me a complete mystery until I heard of the germ theory of putrefaction, when it at once occurred to me that it was only natural that air should be filtered of germs by the air passages, one of whose offices is to arrest inhaled particles of dust, and prevent them from entering the air-cells."

When, in 1869, Tyndall first discovered the remarkably complete absence of dust from exhaled air, particularly toward the end of the expiration, he was engaged in physical researches unrelated to the problem of spontaneous generation. On subsequently encountering Lister's conjecture, noted above, Tyndall recognized that his own observation demonstrated that, in its freedom from dust, air filtered by the deeper cells of the human lungs is "in the precise condition required by Professor Lister's explanation." Thus Tyndall was led to see that his purely physical studies of the optical effects of dust in air might have an important bearing on the biological problem of spontaneous generation.

Lister's most important work in the applied art of surgery was inspired by Pasteur's fundamental studies of fermentation and spontaneous generation. And, as an example of the reciprocal interaction of fundamental and applied science, we see here how Tyndall's fundamental study of spontaneous generation was stimulated by Lister.

> In all cases the correspondence between my experiments and those of Schwann, Schroeder, Pasteur, and Lister in regard to spontaneous generation was perfect. The air which they found inoperative was proved by the luminous beam to be optically pure and therefore germless. Having worked at the subject both by experiment and reflection, on Friday evening, January 21, 1870, I brought it before the members of the Royal Institution. Two or three months subsequently, for sufficient practical reasons, I ventured to direct public attention to the subject in a letter to the *Times*. Such was my first contact with this important question.
>
> This letter, I believe, gave occasion for the first public utterance of Dr. Bastian in relation to this subject. He did me the honour to inform me, as others had informed Pasteur, that the subject 'pertains to the biologist and physician.' He expressed 'amazement' at my reasoning, and warned me that before what I had done could be undone 'much irreparable mischief might be occasioned.' With far less preliminary experience to guide and warn him, the English heterogenist [upholder of the doctrine of spontaneous generation] was far bolder than Pouchet in his experiments, and far more adventurous in his conclusions. With organic infusions he obtained the results of his celebrated predecessor, but he did much more — the atoms and molecules of inorganic liquids passing under his manipulation into those more 'complex chemical compounds,' which we dignify by calling them 'living organisms.'[1] As regards the public who take an interest in such things, and apparently also as regards a large portion of the medical profession, our extremely clever countryman succeeded in restoring the subject to a state of uncertainty similar to that which followed the publication of Pouchet's volume in 1859.

Writing in 1876, Tyndall had offered a fuller view of the lack of decisiveness displayed by Pasteur's 'decisive proofs.' Tyndall remarked that: "Pasteur's labors, which have so long been considered models by most of us, have been subjected to rough handling of late. His reasoning has been criticized, and experiments counter to his have been adduced in such number and variety, and with such an appearance of circumstantial accuracy, as to render the evidence against him overwhelming to many minds. This, I have reason to know, has been the effect wrought, not only upon persons untrained in science, but also upon biologists of eminence both in this country and America. The state of medical opinion in England is correctly described in a recent number of the 'British Medical Journal,' where, in answer to the question, 'In what way is contagium generated and communicated?' we have the reply that, notwithstanding 'an almost incalculable amount of patient labor, the actual results obtained, especially as regards the manner of generation of contagium, have been most disappointing. Observers are even yet at variance whether these minute particles, whose discovery we have just noticed, and other disease-germs, are always produced from like bodies previously existing, or whether they do not, under certain favorable conditions, spring into existence *de novo*.' "

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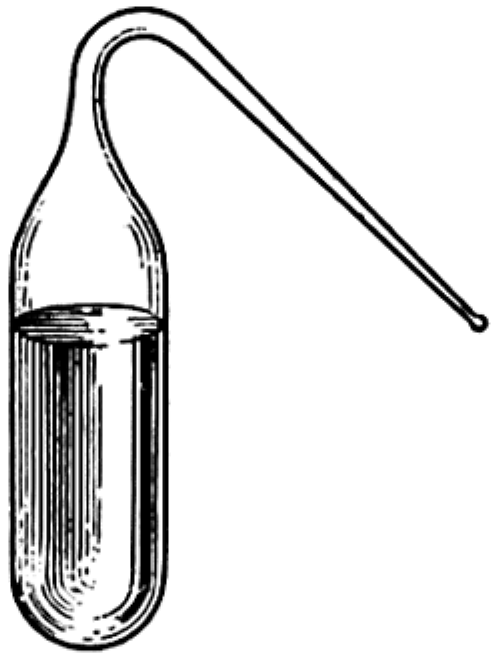
[1] 'It is further held that bacteria or allied organisms are prone to be engendered as correlative products, coming into existence in the several fermentations, just as independently as other less complex chemical compounds.' — Bastian, *Trans. of Pathological Society*, vol. xxvi. 258.

It is desirable that this uncertainty should be removed from all minds, and doubly desirable on practical grounds that it should be removed from the minds of medical men. In the present article, therefore, I propose discussing this question face to face with some eminent and fair-minded member of the medical profession who, as regards spontaneous generation, entertains views adverse to mine. Such a one it would be easy to name; but it is perhaps better to rest in the impersonal. I shall therefore



simply call my proposed co-inquirer my friend. With him at my side, I shall endeavour, to the best of my ability, so to conduct this discussion that he who runs may read and that he who reads may understand.

Let us begin at the beginning. I ask my friend to step into the laboratory of the Royal Institution, where I place before him a basin of thin turnip slices barely covered with distilled water kept at a temperature of 120° Fahr. After digesting the turnip for four or five hours we pour off the liquid, boil it, filter it, and obtain an infusion as clear as filtered drinking water. We cool the infusion, test its specific gravity, and find it to be 1006 or higher — water being 1000. A number of small clean empty flasks, of the shape shown in the figure, are before us. One of them is slightly warmed with a spirit-lamp, and its open end is then dipped into the turnip infusion. The warmed glass is afterwards chilled, the air within the flasks cools, contracts, and is followed in its contraction by the infusion. Thus we get a small quantity of liquid into the flask. We now heat this liquid carefully. Steam is produced, which issues from the open neck, carrying the air of the flask along with it. After a few seconds' ebullition, the open neck is again plunged into the infusion. The steam within the flask condenses, the liquid enters to supply its place, and in this way we fill our little flask to about four-fifths of its volume. This description is typical; we may thus fill a thousand flasks with a thousand different infusions.

I now ask my friend to notice a trough made of sheet copper, with two rows of handy little Bunsen burners underneath it. This trough, or bath, is nearly filled with oil; a piece of thin plank constitutes a kind of lid for the oil-bath. The wood is perforated with circular apertures wide enough to allow our small flask to pass through and plunge itself in the oil, which has been heated, say, to 250° Fahr. Clasped all round by the hot liquid, the infusion in the flask rises to its boiling point, which is not sensibly over 212° Fahr. Steam issues from the open neck of the flask, and the boiling is continued for five minutes. With a pair of small brass tongs, an assistant now seizes the neck near its junction with the flask, and partially lifts the latter out of the oil. The steam does not cease to issue, but its violence is abated. With a second pair of tongs held in one hand, the neck of the flask is seized close to its open end, while with the other hand a Bunsen's flame or an ordinary spirit flame is brought under the middle of the neck. The glass reddens, whitens, softens, and as it is gently drawn out the neck diminishes in diameter, until the canal is completely blocked up. The second pair of tongs with the fragment of severed neck being withdrawn, the flask, with its contents diminished by evaporation, is lifted from the oil-bath perfectly sealed hermetically.

Sixty such flasks filled, boiled, and sealed in the manner described, and containing strong infusions of beef, mutton, turnip, and cucumber, are carefully packed in sawdust, and transported to the Alps. Thither, to an elevation of about 7,000 feet above the sea, I invite my co-inquirer to accompany me. It is the month of July, and the weather is favourable to putrefaction. We open our box at the Bel Alp, and count out fifty-four flasks, with their liquids as clear as filtered drinking water. In six flasks, however, the infusion is found muddy. We closely examine these, and discover that every one of them has had its fragile end broken off in the transit from London. Air has entered the flasks, and the observed muddiness is the result. My colleague knows as well as I do what this means. Examined with a pocket-lens, or even with a microscope of insufficient power, nothing living is seen in the muddy liquid; but regarded with a magnifying power of a thousand diameters or so, what an astonishing appearance does it present! Leeuwenhoek [Antony van Leeuwenhoek (1632–1723), a pioneer in the use of the microscope] estimated the population of a single drop of stagnant water at 500,000,000: probably the population of a drop of our turbid infusion would be this many times multiplied. The field of the microscope is crowded with organisms, some wabbling slowly, others shooting rapidly across the microscopic field. They dart hither and thither like a rain of minute projectiles; they pirouette and spin so quickly round, that the retention of the retinal impression transforms the little living rod into a twirling wheel. And yet the most celebrated naturalists tells us they are vegetables. From the rod-like shape which they so frequently assume, these organisms are called 'bacteria' — a term, be it here remarked, which covers organisms of very diverse kinds.

Has this multitudinous life been spontaneously generated in these six flasks, or is it the progeny of living germinal matter carried into the flasks by the entering air? If the infusions have a self-generative power, how are the sterility and consequent clearness of the fifty-four uninjured flasks to be accounted for? My colleague may urge — and fairly urge — that the assumption of germinal matter is by no means necessary; that the air itself may be the one thing needed to wake up the dormant infusions. We will examine this point immediately. But meanwhile I would remind him that I am working on the exact lines laid down by our most conspicuous heterogenist [Bastian]. He distinctly affirms that the withdrawal of the atmospheric pressure above the infusion favours the production of organisms; and he accounts for their absence in tins of preserved meat, fruit, and vegetables, by the hypothesis that fermentation *has* begun in such tins, that gases *have* been generated, the pressure of which has stifled the incipient life and stopped its further development.[1] This is the new theory of preserved meats. Had Dr. Bastian pierced a tin of preserved meat, fruit, or vegetable under water with the view of testing its truth, he would have found it erroneous. In well-preserved tins he would have found, not an outrush of gas, but an inrush of water. I have noticed this recently in tins which have lain perfectly good for sixty-three years in the Royal Institution. Modern tins, subjected to the same test, yielded the same result. From time to time, moreover, during the last two years, I have placed glass tubes, containing clear infusions of turnip, hay, beef, and mutton, in iron bottles, and subjected them to air-pressures varying from ten to twenty-seven atmospheres — pressures, it is needless to say, far more than sufficient to tear a preserved meat tin to shreds. After ten days these infusions were taken from their bottles rotten with putrefaction and teeming with life. Thus collapses an hypothesis which had no rational foundation, and which could never have seen the light had any well-directed attempt been made to verify it.

Our fifty-four vacuous and pellucid flasks also declare against the heterogenist. We expose them to a warm Alpine sun by day, and at night we suspend them in a warm kitchen. Four of them have been accidentally broken; but at the end of a month we find the fifty remaining ones as clear as at the commencement. There is no sign of putrefaction or of life in any of them. We divide these flasks into two groups of twenty-three and twenty-seven respectively (an accident of counting rendered the division uneven). The question now is whether the admission of air can liberate any generative energy in the infusions. Our next experiment will answer this question and something more. We carry the flasks to a hayloft, and there, with a pair of steel pliers, snip off the sealed ends of the group of three-and-twenty. Each snipping off is of course followed by an inrush of air. We now carry our twenty-seven flasks, our pliers, and a spirit-lamp, to a ledge overlooking the Aletsch glacier, about 200 feet above the hayloft, from which ledge the mountain falls almost precipitously to the north-east for about a thousand feet. A gentle wind blows towards us from the north-east — that is, across the crests and snow-fields of the Oberland mountains. We are therefore bathed by air which must have been for a good while out of practical contact with either animal or vegetable life. I stand carefully to leeward of the flasks, for no dust or particle from my clothes or body must be blown towards them. An assistant ignites the spirit-lamp, into the flame of which I plunge the pliers, thereby destroying all attached germs or organisms. Then I snip off the sealed end of the flask. Prior to every snipping the same process is gone through, no flask being opened without the previous cleansing of the pliers by the flame. In this way we charge our seven-and-twenty flasks with clean vivifying mountain air.

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[1] 'Beginnings of Life,' vol. i. p. 418.

We place the fifty flasks, with their necks open, over a kitchen stove, in a temperature varying from 50° to 90° Fahr., and in three days find twenty-one out of the twenty-three flasks opened on the hayloft invaded by organisms — two only of the group remaining free from them. After three weeks' exposure to precisely the same conditions, *not one of the twenty-seven flasks opened in free air had given way.* No germ from the kitchen air had ascended the narrow necks, the flasks being shaped so as to avoid this contingency. They are still in the Alps, as clear, I doubt not, and as free from life as they were when sent off from London.[1]

What is my colleague's conclusion from the experiment before us? Twenty-seven putrescible infusions, first in vacuo, and afterwards supplied with the most invigorating air, have shown no sign of putrefaction or of life. And as to the others, I almost shrink from asking him whether the hayloft has rendered them spontaneously generative. Is not the inference here imperative that it is not the air of the loft — which is connected through a constantly open door with the general atmosphere — but something contained in the air, that has produced the effects observed? What is this something? A sunbeam entering through a chink in the roof or wall, and traversing the air of the loft, would show it to be laden with suspended dust particles. Indeed the dust is distinctly visible in the diffused daylight. Can *it* have been the origin of the observed life? If so, are we not bound by all antecedent experience to regard these fruitful particles as the germs of the life observed?

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[1] An actual experiment made at the Bel Alp is here described.

. . . . . .

Let us now return to London and fix our attention on the dust of *its* air. Suppose a room in which the housemaid has just finished her work to be completely closed, with the exception of an aperture in a shutter through which a sunbeam enters and crosses the room. The floating dust reveals the track of the light. Let a lens be placed in the aperture to condense the beam. Its parallel rays are now converged to a cone, at the apex of which the dust is raised to almost unbroken whiteness by the intensity of its illumination. Defended from all glare, the eye is peculiarly sensitive to this scattered light. The floating dust of London rooms is organic, and may be burned without leaving visible residue. The action of a spirit-lamp flame upon the floating matter has been elsewhere thus described: —

"In a cylindrical beam which strongly illuminated the dust of our laboratory, I placed an ignited spirit lamp. Mingling with the flame, and round its rim, were seen curious wreaths of darkness resembling an intensely black smoke. On placing the flame at some distance below the beam, the same dark masses stormed upwards. They were blacker than the blackest smoke ever seen issuing from the funnel of a steamer; and their resemblance to smoke was so perfect as to prompt the conclusion that the apparently pure flame of the alcohol-lamp required but a beam of sufficient intensity to reveal its clouds of liberated carbon.

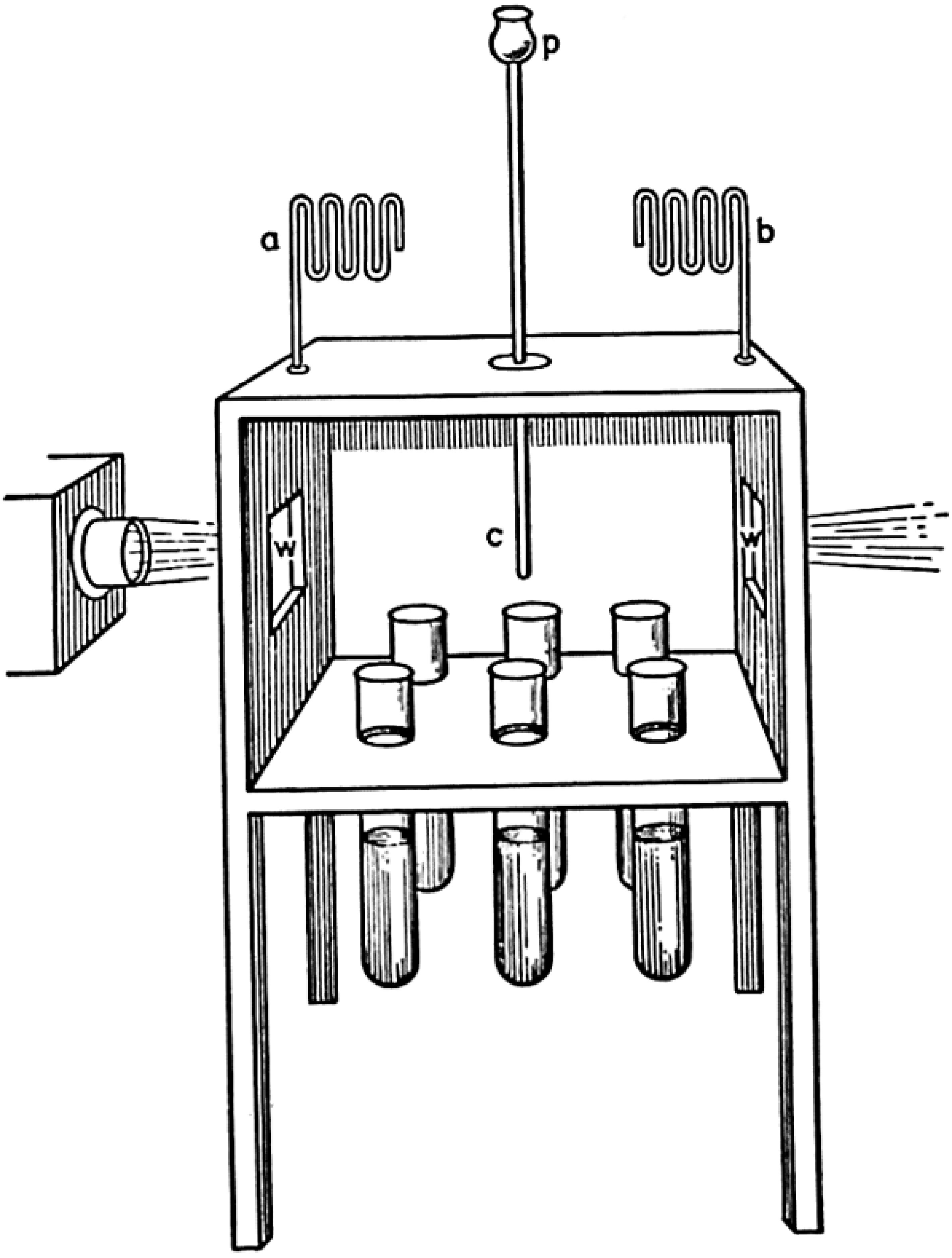
"But is the blackness smoke? This question presented itself in a moment, and was thus answered: A red-hot poker was placed underneath the beam; from it the black wreaths also ascended. A large hydrogen flame, which emits no smoke, was next employed, and it also produced with augmented copiousness those whirling masses of darkness. Smoke being out of the question, what is the blackness? Is it simply that of stellar space; that is to say, blackness resulting from the absence from the track of the beam of all matter competent to scatter its light. When the flame was placed below the beam, the floating matter was destroyed *in situ*; and the heated air, freed from this matter, rose into the beam, jostled aside the illuminated particles, and substituted for their light the darkness due to its own perfect transparency. Nothing could more forcibly illustrate the invisibility of the agent which renders all things visible. The beam crossed, unseen, the black chasm formed by the transparent air, while, at both sides of the gap, the thick-strewn particles shone out like a luminous solid under the powerful illumination."

Supposing an infusion intrinsically barren, but readily susceptible of putrefaction when exposed to common air, to be brought into contact with this unilluminable air, what would be the result? It would never putrefy. It might, however, be urged that the air is spoiled by its violent calcination. Oxygen passed through a spirit-lamp flame is, it may be thought, no longer the oxygen suitable for the development and maintenance of life. We have an easy escape from this difficulty, which is based, however, upon the unproved assumption that the air has been affected by the flame. Let a condensed beam be sent through a large flask or bolthead [a glass vessel having a long, straight neck] containing common air. The track of the beam is seen within the flask — the dust revealing the light, and the light revealing the dust. Cork the flask, stuff its neck with cotton-wool, or simply turn it mouth downwards and leave it undisturbed for a day or two. Examined afterwards with the luminous beam, no track is visible; the light passes through the flask as through a vacuum. The floating matter has abolished itself, being now attached to the interior surface of the flask. Were it our object, as it will be subsequently, to effectually detain the dirt, we might coat that surface with some sticky substance. Here, then, without 'torturing' the air in any way, we have found a means of ridding it, or rather of enabling it to rid itself, of floating matter.

We have now to devise a means of testing the action of such spontaneously purified air upon putrescible infusions. Wooden chambers, or cases, are accordingly constructed, having glass fronts, side-windows, and back-doors. Through the bottoms of the chambers test-tubes pass air-tight; their open ends, for about one-fifth of the length of the tubes, being within the chambers. Provision is made for a free connexion through sinuous channels between the inner and the outer air. Through such channels, though open, no dust will reach the chamber. The top of each chamber is perforated by a circular hole two inches in diameter, closed air-tight by a sheet of india-rubber. This is pierced in the middle by a pin, and through the pinhole is pushed the shank of a long pipette, ending above in a small funnel. The shank also passes through a stuffing box of cotton-wool moistened with glycerine; so that, tightly clasped by the rubber and wool, the pipette is not likely in its motions up and down to carry any dust into the chamber. The annexed figure shows a chamber, with six test-tubes, its side-windows *w w*, its pipette *p c*, and its sinuous channels *a b* which connect the air of the chamber with the outer air.

The chamber is carefully closed and permitted to remain quiet for two or three days. Examined at the beginning by a beam sent through its windows, the air is found laden with floating matter, which in three days has wholly disappeared. To prevent its ever rising again, the internal surface of the chamber was at the outset coated with glycerine. The fresh but putrescible liquid is introduced into the six tubes in succession by means of the pipette. Permitted to remain without further precaution, every one of the tubes would putrefy and fill itself with life. The liquid has been in contact with the dust-laden air outside by which it has been infected, and the infection must be destroyed. This is done by plunging the six tubes into a bath of heated oil and boiling the infusion. The time requisite to destroy the infection depends wholly upon its nature. Two minutes' boiling suffices to destroy some contagia, whereas two hundred minutes' boiling fails to destroy others. After the infusion has been sterilized, the oil-bath is withdrawn, and the liquid, whose putrescibility has been in no way affected by the boiling, is abandoned to the air of the chamber.

With such chambers I tested, in the autumn and winter of 1875–6, infusions of the most various kinds, embracing natural animal liquids, the flesh and viscera of domestic animals, game, fish, and vegetables. More than fifty chambers, each with its series of infusions, were tested, many of them repeatedly. There was no shade of uncertainty in any of the results. In every instance we had, within the chamber, perfect limpidity and sweetness, which in some cases lasted for more than a year — without the chamber, with the same infusion, putridity and its characteristic smells. In no instance was the least countenance lent to the notion that an infusion deprived by heat of its inherent life, and placed in contact with air cleansed of its visibly suspended matter, has any power to generate life anew.



Remembering then the number and variety of the infusions employed, and the strictness of our adherence to the rules of preparation laid down by the heterogenists themselves; remembering that we have operated upon the very substances recommended by them as capable of furnishing, even in untrained hands, easy and decisive proofs of spontaneous generation, and that we have added to their substances many others of our own — if this pretended generative power were a reality, surely it must have manifested itself somewhere. Speaking roundly, I should say that in such closed chambers at least five hundred chances have been given to it, but it has nowhere appeared.

The argument is now to be clenched by an experiment which will remove every residue of doubt as to the ability of the infusions here employed to sustain life. We open the back doors of our sealed chambers, and permit the common air with its floating particles to have access to our tubes. For three months they have remained pellucid and sweet — flesh, fish, and vegetable extracts purer than ever cook manufactured. Three days' exposure to the dusty air suffices to render them muddy, fetid, and swarming with infusorial life. The liquids are thus proved, one and all, ready for putrefaction when the contaminating agent is applied. I invite my colleague to reflect on these facts. How will he account for the absolute immunity of a liquid exposed for months in a warm room to optically pure air, and its infallible putrefaction in a few days when exposed to dust-laden air? He must, I submit, bow to the conclusion that the dust-particles are the cause of putrefactive life. And unless he accepts the hypothesis that these particles, being dead in the air, are in the liquid miraculously kindled into living things, he must conclude that the life we have observed springs from germs or organisms diffused through the atmosphere.

The experiments with hermetically-sealed flasks have reached the number of 940. A sample group of 130 of them were laid before the Royal Society on January 13, 1876. They were utterly free from life, having been completely sterilized by three minutes' boiling. Special care had been taken that the temperatures to which the flasks were exposed should include those previously alleged to be efficient. The conditions laid down by the heterogenist were accurately copied, but there was no corroboration of his results. Stress was then laid on the question of warmth, thirty degrees being suddenly added to the temperatures with which both of us had previously worked. Waiving all protest against the caprice thus manifested, I met this new requirement also. The sealed tubes, which had proved barren in the Royal Institution, were suspended in perforated boxes, and placed under the supervision of an intelligent assistant in the Turkish Bath in Jermyn Street. From two to six days had been allowed for the generation of organisms in hermetically-sealed tubes. Mine remained in the washing-room of the bath for nine days. Thermometers placed in the boxes, and read off twice or three times a day, showed the temperature to vary from a minimum of 101° to a maximum of 112° Fahr. At the end of nine days the infusions were as clear as at the beginning. They were then removed to a warmer position. A temperature of 115° had been mentioned as particularly favourable to spontaneous generation. For fourteen days the temperature of the Turkish Bath hovered about this point, falling once as low as 106°, reaching 116° on three occasions, 118° on one, and 119° on two. The result was quite the same as that just recorded. The higher temperatures proved perfectly incompetent to develop life.

Taking the actual experiment we have made as a basis of calculation, if our 940 flasks were opened on the hayloft of the Bel Alp, 858 of them would become filled with organisms. The escape of the remaining 82 strengthens our case, proving as it does conclusively that not in the air, nor in the infusions, nor in anything continuous diffused through the air, but in *discrete particles*, suspended in the air and nourished by the infusions, we are to seek the cause of life. Our experiment proves these particles to be in some cases so far apart on the hayloft as to permit 10 per cent. of our flasks to take in air without contracting contamination. A quarter of a century ago Pasteur proved the cause of 'so-called spontaneous generation' to be *discontinuous*. [See page 39.] I have already referred to his observation that 12 out of 20 flasks opened on the plains escaped infection, while 19 out of 20 flasks opened on the Mer de Glace escaped. Our own experiment at the Bel Alp is a more emphatic instance of the same kind, 90 per cent. of the flasks opened in the hayloft being smitten, while not one of those opened on the free mountain ledge was attacked.

The power of the air as regards putrefactive infection is incessantly changing through natural causes, and we are able to alter it at will. Of a number of flasks opened in 1876 in the laboratory of the Royal Institution, 42 per cent. were smitten, while 58 per cent. escaped. In 1877 the proportion in the same laboratory was 68 per cent. smitten, to 32 intact. The greater mortality, so to speak, of the infusions in 1877 was due to the presence of hay which diffused its germinal dust in the laboratory air, causing it to approximate as regards infective virulence to the air of the Alpine loft. I would ask my friend to bring his scientific penetration to bear upon all the foregoing facts. They do not prove spontaneous generation to be 'impossible.' My assertions, however, relate not to 'possibilities,' but to *proofs*, and the experiments just described do most distinctly prove the evidence on which the heterogenist relies to be written on waste paper.

My colleague will not, I am persuaded, dispute these results; but he may be disposed to urge that other able and honourable men working at the same subject have arrived at conclusions different from mine. Most freely granted; but let me here recur to the remarks already made in speaking of the experiments of Spallanzani, to the effect that the failure of others to confirm his results by no means upsets their evidence. To fix the ideas, let us suppose that my colleague comes to the laboratory of the Royal Institution, repeats there my experiments, and obtains confirmatory results; and that he then goes to University or King's College, where, operating with the same infusions, he obtains contradictory results. Will he be disposed to conclude that the selfsame substance is barren in Albemarle Street and fruitful in Gower Street or the Strand? His Alpine experience has already made known to him the literally infinite differences existing between different samples of air as regards their capacity for putrefactive infection. And, possessing this knowledge, will he not substitute for the adventurous conclusion that an organic infusion is barren at one place and spontaneously generative at another, the more rational and obvious one that the atmospheres of the two localities which have had access to the infusion are infective in different degrees?

As regards workmanship, moreover, he will not fail to bear in mind, that *fruitfulness* may be due to errors of manipulation, while *barrenness* involves the presumption of correct experiment. It is only the careful worker that can secure the latter, while it is open to every novice to obtain the former. Barrenness is the result at which the conscientious experimenter, whatever his theoretic convictions may be, ought to aim, omitting no pains to secure it, and resorting, only when there is no escape from it, to the conclusion that the life observed comes from no source which correct experiment could neutralize or avoid.

Let us again take a definite case. Supposing my colleague to operate with the same apparent care on 100 infusions — or rather on 100 samples of the same infusion — and that 50 of them prove fruitful and 50 barren. Are we to say that the evidence for and against heterogeny is equally balanced? There are some who would not only say this, but who would treasure up the 50 fruitful flasks as 'positive' results, and lower the evidential value of the 50 barren flasks by labelling them 'negative' results. This, as shown by Dr. William Roberts, is an exact inversion of the true order of the terms positive and negative.[1] Not such, I trust, would be the course pursued by my friend. As regards the 50 fruitful flasks he would, I doubt not, repeat the experiment with redoubled care and scrutiny, and not by one repetition only, but by many, assure himself that he had not fallen into error. Such faithful scrutiny, fully carried out, would infallibly lead him to the conclusion that here, as in all other cases, the evidence in favour of spontaneous generation crumbles in the grasp of the competent inquirer.

The botanist knows that different seeds possess different powers of resistance to heat.[2] Some are killed by a momentary exposure to the boiling temperature, while others withstand it for several hours. Most of our ordinary seeds are rapidly killed, while Pouchet made known to the Paris Academy of Sciences in 1866, that certain seeds, which had been transported in fleeces of wool from Brazil, germinated after four hours' boiling. The germs of the air vary as much among themselves as the seeds of the botanist. In some localities the diffused germs are so tender that boiling for five minutes, or even less, would be sure to destroy them all; in other localities the diffused germs are so obstinate, that many hours' boiling would be requisite to deprive them of their power of germination. The absence or presence of a truss of desiccated hay would produce differences as great as those here described. The greatest endurance that I have ever observed — and I believe it is the greatest on record — was a case of survival after eight hours' boiling.

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[1] See his truly philosophical remarks on this head in the British Medical Journal, 1876, p. 282.

[2] I am indebted to Dr. Thiselton Dyer for various illustrations of such differences. It is, however, surprising that a subject of such high scientific importance should not have been more thoroughly explored. Here the scoundrels who deal in killed seeds might be able to add to our knowledge.

As regards their power of resisting heat, the infusorial germs of our atmosphere might be classified under the following and intermediate heads: Killed in five minutes; not killed in five minutes but killed in fifteen; not killed in fifteen minutes but killed in thirty; not killed in thirty minutes but killed in an hour; not killed in an hour but killed in two hours; not killed in two but killed in three hours; not killed in three but killed in four hours. I have had several cases of survival after four and five hours' boiling, some survivals after six, and one after eight hours' boiling. Thus far has experiment actually reached; but there is no valid warrant for fixing upon even eight hours as the extreme limit of vital resistance. Probably more extended researches (though mine have been very extensive) would reveal germs more obstinate still. It is also certain that we might begin earlier, and find germs which are destroyed by a temperature far below that of boiling water. In the presence of such facts, to speak of *a* death-point of bacteria and their germs would be unmeaning — but of this more anon.

'What present warrant,' it has been asked, 'is there for supposing that a naked, or almost naked, speck of protoplasm can withstand four, six, or eight hours' boiling?' Regarding naked specks of protoplasm I make no assertion. I know nothing about them, save as the creatures of fancy. But I do affirm, not as a 'supposition,' nor an 'assumption,' nor a 'probable guess,' nor as 'a wild hypothesis,' but as a matter of the most undoubted fact, that the spores of the hay bacillus, when thoroughly desiccated [dried] by age, have withstood the ordeal mentioned. [These are the spores that gave Pouchet his apparent evidence of spontaneous generation caused by oxygen]. And I further affirm that these obdurate germs, under the guidance of the knowledge that they *are* germs, can be destroyed by five minutes' boiling, or even less. This needs explanation. The finished bacterium perishes at a temperature far below that of boiling water, and it is fair to assume that the nearer the germ is to its final sensitive condition the more readily will it succumb to heat. Seeds soften before and during germination. This premised, the simple description of the following process will suffice to make its meaning understood.

An infusion infected with the most powerfully resistent germs, but otherwise protected against the floating matters of the air, is gradually raised to its boiling-point. Such germs as have reached the soft and plastic state immediately preceding their development into bacteria are thus destroyed. The infusion is then put aside in a warm room for ten or twelve hours. If for twenty-four, we might have the liquid charged with well-developed bacteria. To anticipate this, at the end of ten or twelve hours we raise the infusion a second time to the boiling temperature, which, as before, destroys all germs then approaching their point of final development. The infusion is again put aside for ten or twelve hours, and the process of heating is repeated. We thus kill the germs *in the order of their resistance*, and finally kill the last of them. No infusion can withstand this process if it be repeated a sufficient number of times. Artichoke, cucumber, and turnip infusions, which had proved specially obstinate when infected with the germs of desiccated hay, were completely broken down by this method of discontinuous heating, three minutes being found sufficient to accomplish what three hundred minutes' continuous boiling failed to accomplish. I applied the method, moreover, to infusions of various kinds of hay, including those most tenacious of life. Not one of them bore the ordeal. These results were clearly foreseen before they were realizd, so that the germ theory fulfils the test of every true theory, that test being the power of prevision. [But how Needham's ghost might have objected to this *repeated* torture of the materials in the infusions!]

When 'naked or almost naked specks of protoplasm' are spoken of, the imagination is drawn upon, not the objective truth of Nature. Such words sound like the words of knowledge where knowledge is really *nil*. The possibility of a 'thin covering' is conceded by those who speak in this way. Such a covering may, however, exercise a powerful protective influence. A thin pellicle of india-rubber, for example, surrounding a pea keeps it hard in boiling water for a time sufficient to reduce an uncovered pea to a pulp. The pellicle prevents imbibition, diffusion, and the consequent disintegration. A greasy or oily surface, or even the layer of air which clings to certain bodies, would act to some extent in a similar way. 'The singular resistance of green vegetables to sterilization,' says Dr. William Roberts, 'appears to be due to some peculiarity of the surface, perhaps their smooth glistening epidermis which prevented complete wetting of their surfaces.' I pointed out in 1876 that the process by which an atmospheric germ is wetted would be an interesting subject of investigation. A dry miscroscope covering-glass may be caused to float on water for a year. A sewing-needle may be similarly kept floating, though its specific gravity is nearly eight times that of water. Were it not for some specific relation between the matter of the germ and that of the liquid into which it falls, wetting would be simply impossible. Antecedent to all development there must be an interchange of matter between the germ and its environment; and this interchange must obviously depend upon the relation of the germ to its encompassing liquid. Anything that hinders this interchange retards the destruction of the germ in boiling water. In my paper, published in the 'Philosophical Transactions' for 1877, I add the following remark:—

"It is not difficult to see that the surface of a seed or germ may be so affected by desiccation and other causes as practically to prevent contact between it and the surrounding liquid. The body of a germ, moreover, may be so indurated by time and dryness as to resist powerfully the insinuation of water between its constituent molecules. It would be difficult to cause such a germ to imbibe the moisture necessary to produce the swelling and softening which precede its destruction in a liquid of high temperature."

However this may be — whatever be the state of the surface, or of the body, of the spores of *Bacillus subtilis* [the hay bacillus], they do as a matter of certainty resist, under some circumstances, exposure for hours to the heat of boiling water. No theoretic scepticism can successfully stand in the way of this fact, established as it has been by hundreds — nay thousands, of rigidly conducted experiments.

We have now to test one of the principal foundations of the doctrine of spontaneous generation as formulated in this country. With this view, I place before my friend and co-inquirer two liquids which have been kept for six months in one of our sealed chambers, exposed to optically pure air. The one is a mineral solution containing in proper proportions all the substances which enter into the composition of bacteria, the other is an infusion of turnip — it might be any one of a hundred other infusions, animal or vegetable. Both liquids are as clear as distilled water, and there is no trace of life in either of them. They are, in fact, completely sterilized. A mutton-chop, over which a little water has been poured to keep its juices from drying up, has lain for three days upon a plate in our warm room. It smells offensively. Placing a drop of the fetid mutton-juice under a microscope, it is found swarming with the bacteria of putrefaction. With a speck of the swarming liquid I inoculate the clear mineral solution and the clear turnip infusion, as a surgeon might inoculate an infant with vaccine lymph. In four-and-twenty hours the transparent liquids have become turbid throughout, and instead of being barren as at first, they are teeming with life. The experiment may be repeated a thousand times with the same invariable result. To the naked eye the liquids at the beginning were alike, being both equally transparent — to the naked eye they are alike at the end, being both equally muddy. Instead of putrid mutton-juice, we might take as a source of infection any one of a hundred other putrid liquids, animal or vegetable. So long as the liquid contains living bacteria, a speck of it communicated either to the clear mineral solution, or to the clear turnip infusion, produces in twenty-four hours the effect here described. [This is the result that Needham's ghost would find most difficult to interpret — particularly so if the experiment were performed with homogeneous cultures or microörganisms (see page 15).]

We now vary the experiment thus: — Opening the back-door of another closed chamber which has contained for months the pure mineral solution and the pure turnip infusion side by side, I drop into each of them a small pinch of laboratory dust. The effect here is tardier than when the speck of putrid liquid was employed. In three days, however, after its infection with the dust, the turnip infusion is muddy, and swarming as before with bacteria. But what about the mineral solution which, in our first experiment, behaved in a manner undistinguishable from the turnip-juice? At the end of three days there is not a bacterium to be found in it. At the end of three weeks it is equally innocent of bacterial life. We may repeat the experiment with the solution and the infusion a hundred times with the same invariable result. Always in the case of the latter the sowing of the atmospheric dust yields a crop of bacteria — never in the former does the dry germinal matter kindle into active life.[1] What is the inference which the reflecting mind must draw from this experiment? Is it not as clear as day that while both liquids are able to feed the bacteria and to enable them to increase and multiply, *after they have been once fully developed*, only one of the liquids is able to develop into active bacteria the germinal dust of the air?

I invite my friend to reflect upon this conclusion; he will, I think, see that there is no escape from it. He may, if he prefers, hold the opinion, which I consider erroneous, that bacteria exist in the air, not as germs but as desiccated organisms. The inference remains, that while the one liquid is able to force the passage from the inactive to the active state, the other is not.

But this is not at all the inference which has been drawn from experiments with the mineral solution. Seeing its ability to nourish bacteria when once inoculated with the living active organism, and observing that no bacteria appeared in the solution after long exposure to the air, the inference was drawn that *neither bacteria nor their germs existed in the air*. Throughout Germany the ablest literature of the subject, even that opposed to heterogeny, is infected with this error; while heterogenists at home and abroad have based upon it a triumphant demonstration of their doctrine. It is proved, they say, by the deportment of the mineral solution that neither bacteria nor their germs exist in the air; hence, if, on exposing a thoroughly sterilized turnip infusion to the air, bacteria appear, they must of necessity have been spontaneously generated. In the words of Dr. Bastian: 'We can only infer that whilst the boiled saline solution is quite incapable of engendering bacteria, such organisms are able to arise *de novo* in the boiled organic infusion.'[2]

I would ask my eminent colleague what he thinks of this reasoning now? The *datum* is — 'A mineral solution exposed to common air does not develop bacteria;' the *inference* is — 'Therefore if a turnip infusion similarly exposed develop bacteria, they must be spontaneously generated.' The inference, on the face of it, is an unwarranted one. But while as matter of logic it is inconclusive, as matter of fact it is chimerical. London air is as surely charged with the germs of bacteria as London chimneys are with smoke. The inference just referred to is completely disposed of by the simple question: 'Why, when your sterilized organic infusion is exposed to optically pure air, should this generation of life *de novo* utterly cease? Why should I be able to preserve my turnip-juice side by side with your saline solution for the three hundred and sixty-five days of the year, in free connexion with the general atmosphere, on the sole condition that the portion of that atmosphere in contact with the juice shall be visibly free from floating dust, while three days' exposure to that dust fills it with bacteria?' Am I over-sanguine in hoping that as regards the argument here set forth he who runs may read, and he who reads may understand?

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[1] This is the deportment of the mineral solution as described by others. My own experiments would lead me to say that the development of the bacteria, though exceedingly slow and difficult, is not impossible. [Investigations in the 20th century have shown that minute quantities of certain organic compounds must be present to support the growth of microörganisms. Whether these would be introduced by the dust would be a matter of accident; they would always be present in a drop of mutton juice.]

[2] Proceedings of the Royal Society, vol. xxi. p. 130.

We now proceed to the calm and thorough consideration of another subject, more important if possible than the foregoing one, but like it somewhat difficult to seize by reason of the very opulence of the phraseology, logical and rhetorical, in which it has been set forth. The subject now to be considered relates to what has been called 'the death-point of bacteria.' Those who happen to be acquainted with the modern English literature of the question will remember how challenge after challenge has been issued to panspermatists in general, and to one or two home workers in particular, to come to close quarters on this cardinal point. [The panspermatists were the upholders of the doctrine of the almost universal diffusion of germs, and thus were the opponents of the doctrine of spontaneous generation.] It is obviously the stronghold of the English heterogenist. 'Water,' he says, 'is boiling merrily over a fire when some luckless person upsets the vessel so that the heated fluid exercises its scathing influence upon an uncovered portion of the body — hand, arm, or face. Here, at all events, there is no room for doubt. Boiling water unquestionably exercises a most pernicious and rapidly destructive effect upon the living matter of which we are composed.' And lest it should be supposed that it is the high organization which, in this case, renders the body susceptible to heat, he refers to the action of boiling water on the hen's egg to dissipate the notion. 'The conclusion,' he says, 'would seem to force itself upon us that there is something intrinsically deleterious in the action of boiling water upon living matter — whether this matter be of high or of low organization.' Again, at another place: 'It has been shown that the briefest exposure to the influence of boiling water is destructive of all living matter.'

The experiments already recorded plainly show that there is a marked difference between the dry bacterial matter of the air, and the wet, soft, and active bacteria of putrefying organic liquids. The one can be luxuriantly bred in the saline solution, the others refuse to be born there, while both of them are copiously developed in a sterilized turnip infusion. Inferences, as we have already seen, founded on the deportment of the one liquid cannot with the warrant of scientific logic be extended to the other. But this is exactly what the heterogenist has done, thus repeating as regards the death-point of bacteria the error into which he fell concerning the germs of the air. Let us boil our muddy mineral solution with its swarming bacteria for five minutes. In the soft succulent condidition in which they exist in the solution not one of them escapes destruction. The same is true of the turnip infusion if it be inoculated with the living bacteria only — the aërial dust being carefully excluded. In both cases the dead organisms sink to the bottom of the liquid, and without re-inoculation no fresh organisms will arise. But the case is entirely different when we inoculate our turnip infusion with the desiccated germinal matter afloat in the air.

The 'death-point' of bacteria is the maximum temperature at which they can live, or the minimum temperature at which they cease to live. If, for example, they survive a temperature of 140°, and do not survive a temperature of 150°, the death-point lies somewhere between these two temperatures. Vaccine lymph [for smallpox vaccination], for example, is proved by Messrs. Braidwood and Vacher to be deprived of its power of infection by brief exposure to a temperature between 140° and 150° Fahr. This may be regarded as the death-point of the lymph, or rather of the particles diffused in the lymph, which constitute the real contagium. If no time, however, be named for the application of the heat, the term 'death-point' is a vague one. An infusion, for example, which will resist five hours' continuous exposure to the boiling temperature, will succumb to five days' exposure to a temperature 50° Fahr. below that of boiling. The fully developed soft bacteria of putrefying liquids are not only killed by five minutes' boiling, but by less than a single minute's boiling — indeed, they are slain at about the same temperature as the vaccine. The same is true of the plastic, active bacteria of the turnip infusion.[1]

But, instead of choosing a putrefying liquid for inoculation, let us prepare and employ our inoculating substance in the following simple way: — Let a small wisp of hay, desiccated by age, be washed in a glass of water, and let a perfectly sterilized turnip infusion be inoculated with the washing liquid. After three hours' continuous boiling the infusion thus infected will often develop luxuriant bacterial life. Precisely the same occurs if a turnip infusion be prepared in an atmosphere well charged with desiccated hay-germs. The infusion in this case infects itself without special inoculation, and its subsequent resistance to sterilization is often very great. On the 1st of March last I purposely infected the air of our laboratory with the germinal dust of a sapless kind of hay mown in 1875. Ten groups of flasks were charged with turnip infusion prepared in the infected laboratory, and were afterwards subjected to the boiling temperature for periods varying from 15 minutes to 240 minutes. Out of the ten groups only one was sterilized — that, namely, which had been boiled for four hours. Every flask of the nine groups which had been boiled for 15, 30, 45, 60, 75, 90, 105, 120, and 180 minutes, respectively, bred organisms afterwards. The same is true of other vegetable infusions. On the 28th of February last, for example, I boiled six flasks, containing cucumber infusion prepared in an infected atmosphere, for periods of 15, 30, 45, 60, 120, and 180 minutes. Every flask of the group subsequently developed organisms. On the same day, in the case of three flasks, the boiling was prolonged to 240, 300, and 360 minutes; and these three flasks were completely sterilized. Animal infusions, which under ordinary circumstances are rendered infallibly barren by five minutes' boiling, behave like the vegetable infusions in an atmosphere infected with hay-germs. On the 30th of March, for example, five flasks were charged with a clear infusion of beef and boiled for 60 minutes, 120 minutes, 180 minutes, 240 minutes, and 300 minutes respectively. Every one of them became subsequently crowded with organisms, and the same happened to a perfectly pellucid mutton infusion prepared at the same time. The cases are to be numbered by hundreds in which similar powers of resistance were manifested by infusions of the most diverse kinds.

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[1] In my paper in the Philosophical Transactions for 1876, I pointed out and illustrated experimentally the difference, as regards rapidity of development, between water-germs and air-germs; the growth from the already softened water-germs proving to be practically as rapid as from developed bacteria. This preparedness of the germ for rapid development is associated with its preparedness for rapid destruction.

In the presence of such facts I would ask my colleague whether it is necessary to dwell for a single instant on the one-sidedness of the evidence which led to the conclusion that all living matter has its life destroyed by 'the briefest exposure to the influence of boiling water.' An infusion proved to be barren by six months' exposure to moteless air maintained at a temperature of 90° Fahr., when inoculated with full-grown active bacteria, fills itself in two days with organisms so sensitive as to be killed by a few minutes' exposure to a temperature much below that of boiling water. But the extension of this result to the desiccated germinal matter of the air is without warrant or justification. This is obvious without going beyond the argument itself. But we have gone far beyond the argument, and proved by multiplied experiment the alleged destruction of all living matter by the briefest exposure to the influence of boiling water to be a delusion. The whole logical edifice raised upon this basis falls therefore to the ground; and the argument that bacteria and their germs, being destroyed at 140°, must, if they appear after exposure to 212°, be spontaneously generated, is, I trust, silenced for ever.

Through the precautions, variations, and repetitions observed and executed with the view of rendering its results secure, the separate vessels employed in this inquiry have mounted up in two years *to nearly ten thousand*.

Besides the philosophic interest attaching to the problem of life's origin, which will be always immense, there are the practical interests involved in the application of the doctrines here discussed to surgery and medicine. The antiseptic system, at which I have already glanced, illustrates the manner in which beneficent results of the gravest moment follow in the wake of clear theoretic insight. Surgery was once a noble art; it is now, as well, a noble science. Prior to the introduction of the antiseptic system, the thoughtful surgeon could not have failed to learn empirically that there was something in the air which often defeated the most consummate operative skill. That something the antiseptic treatment destroys or renders innocuous. At King's College Mr. Lister operates and dresses while a fine shower of mixed carbolic acid and water, produced in the simplest manner, falls upon the wound, the lint and gauze employed in the subsequent dressing being duly saturated with the antiseptic. At St. Bartholomew's Mr. Callender employs the dilute carbolic acid without the spray; but, as regards the real point aimed at — the preventing of the wound from becoming a nidus [nest, focus] for the propagation of septic bacteria — the practice in both hospitals is the same. Commending itself as it does to the scientifically trained mind, the antiseptic system has struck deep root in Germany.

Had space allowed, it would have given me pleasure to point out the present position of the 'germ theory' in reference to the phenomena of infectious disease, distinguishing arguments based on analogy — which, however, are terribly strong — from those based on actual observation. I should have liked to follow up the account I have already given[1] of the truly excellent researches of a young and an unknown German physician named Koch, on splenic fever [anthrax], by an account of what Pasteur has recently done with reference to the same subject. Here we have before us a living contagium of the most deadly power, which we can follow from the beginning to the end of its life cycle. We find it in the blood or spleen of a smitten animal in the state say of short motionless rods. When these rods are placed in a nutritive liquid on the warm stage of the microscope, we soon see them lengthening into filaments which lie, in some cases, side by side, forming in others graceful loops, or becoming coiled into knots of a complexity not to be unravelled. We finally see those filaments resolving themselves into innumerable spores, each with death potentially housed within it, yet not to be distinguished microscopically from the harmless germs of *Bacillus subtilis*. The bacterium of splenic fever is called *Bacillus Anthracis*. This formidable organism was shown to me by M. Pasteur in Paris last July. His recent investigations regarding the part it plays pathologically certainly rank amongst the most remarkable labours of that remarkable man. Observer after observer had strayed and fallen in this land of pitfalls, a multitude of opposing conclusions and mutually destructive theories being the result. In association with a younger physiological colleague, M. Joubert, Pasteur struck in amidst the chaos, and soon reduced it to harmony. They proved, among other things, that in cases where previous observers in France had supposed themselves to be dealing solely with splenic fever, another equally virulent factor was simultaneously active. Splenic fever was often overmastered by septicaemia, and results due solely to the latter had been frequently made the ground of pathological inferences regarding the character and cause of the former. Combining duly the two factors, all the previous irregularities disappeared, every result obtained receiving the fullest explanation. On studying the account

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[1] 'Fortnightly Review,' November, 1876. [This article is reproduced in Case 6.]

> of this masterly investigation, the words wherewith Pasteur himself feelingly alludes to the difficulties and dangers of the experimenter's art came home to me with especial force: 'J'ai tant de fois éprouvé que dans cet art difficile de l'expérimentation les plus habiles bronchent à chaque pas, et que l'interprétation des faits n'est pas moins périlleuse.'[1]

Pointing to the important advances made by Pasteur, Lister and Koch, Tyndall stresses the great fruitfulness of research predicated on the hypothesis that (spontaneous generation being impossible) disease germs must always have living precursors. The striking successes achieved through these studies afforded strong support to Pasteur's opinion, and by the end of the nineteenth century the impossibility of spontaneous generation was finally established as a valuable working principle. Many philosophers of science would say that every scientific theory must establish its claim by its service as a guide to fruitful action. This would be as true of the law of definite proportions, or the atomic theory, or the law of the conservation of mass, as of the case at hand.

There is, however, one point in the present case that needs emphasis. Pasteur and Tyndall were endeavoring to establish a principle that was cast in a negative form. This made it difficult for them to *prove* their point by any finite group of experiments. Even today no one can categorically deny that somewhere on earth living organisms are being produced from nonliving matter. The situation was well summed up by Pasteur when, in speaking of Bastian's work, he said: "More fortunate than inventors of perpetual motion, the champions of spontaneous generation will, for a long time yet, be privileged to arouse the attention of the scientific world. In the mathematical sciences it can be demonstrated that a given proposition is not and could not be so, but the sciences of nature are not so well devised. Mathematicians can reject unread all memoirs concerned with the squaring of the circle or perpetual motion. The question of spontaneous generation, on the contrary, is always capable of inflaming public opinion. For, in the present state of science, it is impossible to prove a priori that there can be no self-creation of life apart from the preëxistence of similar living forms." Only a slow development — a gradual elimination of one alleged case of spontaneous generation after another, an ever-widening demonstration of the fruitfulness of Pasteur's hypothesis, an intensive study of the propagation of pure cultures (see p. 15) — has led to the general conviction of the truth of this hypothesis, on which all modern microbiology is based. One can at least say that there have been obtained no reproducible results that are not far better explained in terms of

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[1] Comptes-Rendus, lxxxiii, p. 177.

Pasteur's hypothesis than in terms of the doctrine of spontaneous generation.

It may be asked: If spontaneous generation is impossible, how then did life originate? Such a question leads one into the study of the ancient past, and a consideration of what evidence we can hope to obtain of events that occurred so long ago and over such vast periods of time. As Pasteur often pointed out, such studies are wholly distinct from his own investigations of the possible occurrence of spontaneous generation of life in the modern world. Those who are interested in the remote origins of life on earth will find various possibilities in the hypotheses currently under discussion by geologists and others concerned with the ancient history of this planet.

SUGGESTED READING

Louis Pasteur, Free Lance of Science, by René J. Dubos (Little, Brown, Boston, 1950). An excellent biography of Pasteur which is for the lay reader a good introduction to the methods of experimental microbiology, written by a man who is himself a leader in this field of experimentation. Chapter VI deals with the controversy as to spontaneous generation; Chapter XIII, "Mechanism of Discoveries," is especially recommended to those who wish to understand the nature of scientific investigations.

The Life of Pasteur, by René Vallery-Radot, translated by Mrs. R. L. Devonshire (Doubleday, Doran, New York, 1928). This is an intimate account of Pasteur's life, arranged chronologically and written by his son-in-law and co-worker. Chapter II to Chapter V, inclusive, provide the background for the scientific work on fermentation and spontaneous generation.

Pasteur's Study of Fermentation, edited by J. B. Conant (Harvard University Press, Cambridge, 1952; Harvard Case Histories in Experimental Science, No. 6). A study of Pasteur's work on fermentation can be combined to good advantage with a study of the controversy in regard to spontaneous generation.

The History of Bacteriology, by William Bulloch (Oxford University Press, London, 1938). Chapter IV, on "Spontaneous Generation and Heterogenesis," provides a full and detailed account of the subject.